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North American Clinical Trials Network (NACTN) for Treatment of Spinal Cord Injury: A Consortium of Military, Veterans Administration, and Civilian Hospitals

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13. SUPPLEMENTARY NOTES

14. ABSTRACT Four hundred forty-one acutely injured patients have been enrolled in the NACTN data registry as of 1/6/2011; enrollment continues at all clinical centers. 24 acutely injured patients have been enrolled in the Riluzole Phase I safety study; to-date, no adverse reactions to the drug have been reported. NACTN has reorganized itself and now operates under a formally adopted Governance Manual and Policies & Procedures and works through established committees; oversight for all NACTN operations is vested in its Executive Committee. Under the aegis of its Neurological Outcomes Committee (NOA), three contracts have been issued to develop and finalize three new outcome instruments. A book chapter on NACTN is in press; the network has submitted its first manuscript for publication; two manuscripts are in preparation by Dr. Diana Chow and the Publications Committee is exploring a NACTN focus issue with J Neurosurgery. NACTN's Treatment Strategy Selection Committee is recommending consideration of a Phase III randomized, controlled trial of Riluzole. Several NACTN investigators met with investigators from the European EM-SCI network and agreed to begin the process of data sharing, which will also include data from the STASCIS trial and from the Reeve Foundation's NeuroRecovery Network. Dr.Grossman and Colonel Randall McCafferty have discussed San Antonio Military Medical Center (SAMMC joining NACTN, which they anticipate can be finalized during Q1 2011.

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INTRODUCTION: In 2009, the Reeve Foundation released its survey conducted by the University of New Mexico, "One Degree of Separation: Paralysis and Spinal Cord Injury in the United States." Heretofore, it had been thought there were approximately 250,000 living Americans who had a suffered a spinal cord injury SCI). According to the survey however, some 5,596,000 individuals are paralyzed, 1.275 million of them from spinal cord injury (Cahill et al., 2009). Lifetime care costs for a 25 year old with a high cervical SCI are estimated to be 3 million dollars (Spinal Cord Information, 2008). Clearly the imperative to develop effective therapies for these individuals is all the more acute in light of the new numbers. Spinal cord patients are living near-normal life spans now, thanks to vastly improved medical care and rehabilitation but as of this writing, not a single intervention, acute or chronic, has been successfully tested in a rigorous randomized, controlled trial and brought to clinical application. Never has the need for the North American Clinical Trials Network (NACTN) been greater.

NACTN is the only established standing network for spinal cord injury clinical trials in North America. It was created in 2004 by the Christopher Reeve Foundation (CRF) and a consortium of university neurosurgical departments. The U.S. Army Medical Research and Materiel Command of the Department of Defense has supported NACTN since 2006.

The Network's mission is to carry out clinical trials of the comparative effectiveness of new therapies for spinal cord injury using an established consortium of neurosurgery departments at university-affiliated medical center hospitals with medical, nursing and rehabilitation personnel who are skilled in the evaluation and management of SCI. To-date, NACTN has developed data collection instruments to characterize the severity of the initial injury and the course of recovery and created a Data Management and Statistical Coordinating Center, a database of the natural history of SCI and a Pharmacological Center. NACTN has standardized data collection and reporting, and in a major accomplishment during this report period, it has established a committee-based infrastructure that codifies its administration and operations and leverages the skills and expertise of its personnel.

The Executive Committee provides governance and addresses long-term issues critical to the goals and objectives of NACTN; Standing Committees [Data Management, Publications, Treatment Strategy Selection and Neurological Outcome Assessments (NOA)] develop Committee policies and procedures and take leadership responsibility in meeting Committee goals.

BODY: The following tasks have been addressed during the contract period June 1, 2009 – December 31, 2010:

1. **Enroll patients with SCI to expand NACTN's statistical model of the functional outcomes of SCI that are stratified and characterized by neurological, physiological and radiological parameters. Goal: 400 patients throughout the network. Status:**

   The Data Registry, a core function of the North American Clinical Trials Network (NACTN), serves two vital purposes. The first is to provide a statistical and scientific platform to develop the data, logistics and collaborations necessary to conduct Phase I and Phase II clinical trials of emerging neuroprotective and neuroregenerative therapies, particularly those that can be administered in the very early stages of injury. A second and equally important purpose is to develop high quality, standardized, and validated acute care and follow-up data on a representative national sample of male and female adult patients who have suffered a spinal cord injury with neurological deficits. This acute care and follow-up data are an invaluable and unique resource needed to characterize the trajectory (natural history) of individuals who have suffered a spinal cord injury.

   All data are collected prospectively starting at the time of admission to a NACTN clinical center. The registry data includes extensive demographic information, past medical history, pre-injury medication use, circumstances of injury, time of injury, and the time of arrival to the treating NACTN hospital. Further detail is elicited about the condition of the patient on arrival and includes a clinical evaluation,

measurement of state of consciousness with the Glasgow Coma Scale (GCS) and of associated injuries with the Abbreviated Injury Scale. The American Spinal Injury Association impairment scale (AIS) is scored on admission and at key times throughout the patients' hospital and post-hospital course. All examiners received training on performing the AIS examination and study procedures. Data are also collected on radiographic findings, non-operative and operative treatments, timing of treatments, and perioperative complications. Discharge AIS score, and the type of facility to which the patient was transferred are recorded in the discharge form. After acute care discharge, Long-term follow-up is scheduled at approximate intervals of six and twelve months after discharge. The follow-up registry protocol includes: the AIS Impairment Scale, and where appropriate, the Functional Independence Measure FIM™, the Spinal Cord Independence Measure (SCIM), and the Walking Index for Spinal Cord Injury (WISCI) evaluations.

Currently there are nine clinical centers participating in the NACTN SCI Registry. .As of December 31, 2010, a total of 441 patients were enrolled into the NACTN Data Registry.

During the period 6/1/2009 – 12/31/2010, 137 new SCI patients were enrolled in the Registry. Also during this time period the Data Management Center (DMC) processed 100 six-month and 66 twelve-month follow-ups for previously enrolled patients. The DMC data quality control program (manual and electronic) was also expanded to accommodate the expanded number of clinical centers and the increased data traffic. The Methodist Hospital Coordinating Center (TMH-CC) and DMC staff also provides training to clinical coordinators at the nine NACTN centers to ensure proper data acquisition and adherence to protocol and data submission procedures.

An overview of the DMC data algorithms developed and flow of manual and computer processing is given in Appendix A. The DMC also developed a system for sharing registry data with NACTN investigators and others approved for data access. Data are provided in the format requested by a user and are provided with a either a de-identified data file or requested tabulations. In the period 6/1/2009 – 12/31/2010 data files and tabulations have been provided to three NACTN investigators (Drs Aarabi, Guest, and Wilson). Registry tabulations are also in the process of being provided to Dr. Michael Wang, University of Miami, to support his NIH-NINDS research application for a clinical trial evaluating hypothermia for the treatment of acute traumatic spinal cord injury.

The tables in Appendix B provide a profile of SCI cases currently in the registry database. As of 12/31/2010, 792 SCI patients have been contacted for permission to acquire prospective, standardized acute care treatment data and quarterly follow-up data for up to one-year after acute care discharge. Slightly more than one-half (56%) of all patients screened for the registry enrollment consented to participate (Table 1). The following text summarizes the current registry database information for 407 of 441 registry patients with the remaining records of 34 patients under quality control review for subsequent inclusion in the database.

Eighty percent of the patients are male, 76% white, and the median age at injury was 43 years for all patients.(Table 2). The leading circumstances of injury were falls (36%), motor vehicle accidents (31%), motorcycle and off-road vehicle accidents (10.0%), recreation accidents (12%), and 6% were due to assault (Table 3). Approximately 58% of all SCI patients arrived by EMS directly to a NACTN clinical center from the scene of injury with a median arrival time of 1.1 hours. Of patients transferred from an intermediate hospital, the arrival time post-injury was 8.9 hours.

The distribution of AIS severity of patients with neurological deficit at admission was AIS A (34.9%), AIS B (13.3%), AIS C (12.0%), AIS D (24.8%), AIS E (8.8%) and AIS was not available for 6.1% of the 407 SCI cases (Table 4).

Of the 407 cases, 57% had at least one mild, moderate or severe complication, and 20.4% had four or more complications (Table 5). Of the total number of complications ascertained (1,079), pulmonary, infections, hematologic, and cardiac complications accounted for 76% of all complications (Table 6). The in-hospital death rate was 14/406 = 0.034 (3.4%) (one case is currently hospitalized).

The vast majority of SCI injuries were blunt injuries or crushing injuries (95%), but 4% were penetrating SCI injuries, primarily bullet injuries. Of the 407 patients, 75% sustained cervical injuries and 18% thoracic injuries (Table 7).

Surgical and corticosteroid treatments are summarized in Tables 8 and 9. About 90% of patients evaluated as AIS A at admission were surgically treated, with decreasing proportions of surgery for less severe AIS grades. Decompression was achieved and confirmed by radiology in approximately 80% of all surgeries. Two-thirds of AIS A through AIS C patients received steroids.

Length of acute care hospitalization and discharge status is summarized in Table 10. For 406 SCI patients, approximately 45% had a length of stay exceeding two weeks. For AIS A patients, the median hospital stay was approximately 19 days. More than two-thirds of the SCI patients were discharged to either a rehabilitation hospital (68%) and 9% were transitioned to long-term acute care. Thirty-nine patients were discharged as either partial or complete ventilator dependent. A rehabilitation program was initiated for 81% of the patients prior to discharge from acute care.

Table 11 contrasts the AIS grades at admission to the AIS grades at hospital discharge for 365 SCI patients for whom complete data is currently available. Notable is that 88% of patients with a grade of AIS A at admission remained AIS A at discharge. Although there was improvement within each AIS grade, the improvement in AIS grade at the time of hospital discharge from acute care was quite modest.

An important milestone was achieved in the registry during 6/1/2009 – 12/31/2010. The registry now has more than 400 case records of acute traumatic SCI and is now poised to become a national resource for SCI research. The case numbers are sufficiently large to have adequate statistical precision for the study of acute care complications and their relationship to outcome at 6-months and 12-months.

Acute Complications Manuscript: Utilize the Registry data to develop a manuscript for publication that describes the incidence, type, and severity of acute care complications.

A considerable amount of DMC staff time during the period 6/1/2009 – 12/31/2010 was devoted to developing the data analysis for the manuscript, Grossman, R.G. et al. (2011). “Incidence and Severity of Acute Care Complications after Spinal Cord Injury.” This manuscript has been completed and was submitted to the Journal of Neurotrauma for publication. A copy of the manuscript is included with this report.

Safety and Pharmacokinetics of Riluzole in Patients with Traumatic Acute Spinal Cord Injury

During the period 6/1/2009 – 12/31/2010, TMH-CC devoted much time to facilitating the regulatory processes at all NACTN clinical sites and at the data management and pharmacological centers, including IRB submission, queries and approvals by the DOD HRPO ORP. Standardized forms and site start-up documents were developed. Master regulatory binders were created and are maintained at each clinical site with a master file at TMH-CC.

TMH developed a Procedural Instructional Guide (PIG, submitted with an earlier quarterly report); this is utilized by the study coordinators and investigators as a check list and step-by-step guide explaining the exact research activities to be conducted from the time of subject enrollment to the final six-month follow-up visit.

TMH-CC, in collaboration with DMC staff and NACTN clinical investigators, developed the data protocol and data collection forms (included with earlier quarterly reports) for the Riluzole safety trial. The data protocol developed includes 14 data collection forms tracking the clinical course and outcomes (safety and complications) starting at the time a patient consents to enter the safety trial and records patient outcomes from the time of acute care through hospital discharge, including follow-up visits at 6-weeks, 3-months and six-months post-injury.

DMC staff also developed and tested a data acquisition system for the Riluzole safety trial. The data system is capable of high levels of data quality control, confidentiality protection and also allows for the efficient merging of pharmacokinetic information with clinical information. An overview of the system is given in Appendix C. The data protocol and data system proposed by the DMC was approved by the University of Texas Health Science Center at Houston Institutional Review Board on 6/18/2010 and reviewed and approved by the USAMRC, Office of Research Protections on 09/08/2010.

TMH-CC and DMC staff trained and developed logistic and communication links with all of the clinical coordinators at each participating NACTN clinical site and assisted the Clinical Coordinating Center in Regulatory issues and various administrative data tasks.

During the maintenance phase of the study, TMH-CC has assisted all clinical sites pertaining to recruitment, enrollment and protocol issues. A 24-hour helpline has been provided. Monthly conference calls are conducted for all key personnel and minutes are posted on the NACTN ftp site, which is accessible by all NACTN personnel.

TMH-CC has conducted routine site management functions and has assisted the Reeve Foundation with sponsor activities. On-site monitoring has been done after the enrollment of the first subject at each site to insure protocol procedures are being strictly followed. On-site activities and identification of actionable items are addressed during these monitoring visits. Monitoring visit reports are prepared for the principal investigator, documenting pertinent points and action items. Subsequent follow-up communications are documented.

Riluzole Trial Enrollment

The target enrollment for the Riluzole Safety trial is 36 patients. The first case was entered in April, 2010. Currently 24 patients have enrolled in this clinical investigation. As of 12/31/2010, the DMC has processed case records for 20 patients enrolled in the safety trial and these patients are described below.

Five tables in Appendix D provide a profile of the 20 patients enrolled (and processed by the DMC) in the Riluzole trial as of 12/31/2010. The patients described were enrolled by five NACTN centers.

The trial protocol requires rapid evacuation of a patient to a NACTN Clinical Center and initiation of Riluzole treatment within 12 hours of injury. Table 1 demonstrates that all patients arrived at a NACTN center within seven hours of injury, and Riluzole treatment was initiated for all 20 patients within 5.5 hours and 12 hours post-injury.

Tables 2 and 3 summarize the demographics and circumstances of injury. The 20 enrolled patients are all male and range in age from 18 to 67 years of age with a median age of 26 years. The ethnic distribution is White (60%), African-American (25%), and Asian/other (15%). Motor vehicle accidents were the leading cause of SCI injury (40%) followed by falls (25%) and diving accidents (15%). Two SCI injuries were attributed to assaults but neither was a penetrating injury. This relatively young SCI population reported few pre-existing health problems; 4 cases of hypertension, 1 case of Diabetes, and 3 cases with prior psychiatric or mental health problems. Eight of the 20 patients were current or former smokers.

The distribution of AIS severity of the 20 patients was AIS A (45%), AIS B (30%), and AIS C (25%). Half of the patients were classified as tetra incomplete and 30% were classified as para complete (Table 4). Table 5 lists the surgeries and corticosteroid use received by the 20 patients. Posterior or anterior + posterior surgery were the most frequent surgeries (70%). Two patients did not receive surgery and there are two cases where surgery has not yet been reported to the DMC. Corticosteroid use was relatively low with only 5 (25%) receiving steroids. Two cases are pending submission to the DMC on steroid use.

All 20 patients completed the prescribed 15 day doses of Riluzole with only minor and inconsequential deviations from the specified protocol dose schedule.

The Riluzole trial is progressing as planned with only minor but correctable problems in data collection. Based on the 2010, it is projected that the target of 36 cases will be achieved at the latest by early fall 2011.

2. **Expansion of Phase I baseline assessment research protocols for hospitals joining NACTN, working with the United States Army Medical Research and Materiel Command (USAMRMC) Office of Research Protections (ORP), Human Research Protection Office (HRPO) and local IRBs. Status:**
   All participating NACTN clinical sites (the exception is Walter Reed Army Medical Center) have received the required institutional and DOD regulatory approvals for the Riluzole safety study and are actively recruiting subjects for it.

3. **Enroll patients with SCI to expand NACTN's statistical model of the functional outcomes of SCI that are stratified and characterized by neurological, physiological and radiological parameters. Goal: 200 patients throughout the network. Status:**

   As of December 31, 2010, a total of 441 patients have been enrolled into the NACTN data registry. See #1, above.

4. **Expansion of NACTN to include military, Veterans Administration and additional civilian hospitals. Status:**
   NACTN's ability to expand beyond its present nine clinical centers was severely limited during this report period due to the delay in authorization and/or release of funds by the Department of Defense. Essentially, the Reeve Foundation was forced to seek modifications to allow funds earmarked for expansion and/or other tasks to be reallocated to insure support for NACTN's centers. These reallocations effectively enabled these centers to consent patients, as appropriate, into the data registry and the Riluzole clinical trial but they meant that no new military or civilian sites were recruited. BAA2010 funds, released in July 2010, include support for two new centers.

5. **Characterize the biomechanical, anatomical and neurological differences between military and civilian injuries and differences in their outcomes. Status:**
   This particular task will be initiated when Walter Reed Army Medical Center enrolls a sufficient number of patients into the NACTN data registry. At this writing, 12 have been enrolled.

   A retrospective component was added to collect spinal cord injury and initial outcome data from WRAMC patients for the period 1 January 2003 to 23 March 2008. Data collected will be matched to the data collected in the prospective portion of the study, with the exception of dates which will be replaced with a "time since injury" calculation. Approval for this retrospective portion was received 5 August 2010. Since then, a new clinical coordinator, Vicky Miskovsky, has been brought onto the WRAMC team and the further addition of Kim Clark, PA-C, a study coordinator, is pending review and approval.

6. **Expand the Data Management Center at the University of Texas School of Public Health to incorporate the increased numbers of patients in the study. Status:**
   Our last quarterly report, submitted 9-2010, documented those expansion activities which allow the Data Management Center to effectively fulfill its responsibilities with respect to all data related to NACTN's registry and initial Phase I Riluzole safety study. There are no additional expansion activities to-date and the DMC report at #1, above, details data flow for both the registry and the clinical trial.

7. **Further validate quantitative measurements to assess neurological recovery, including the Graded Refined Assessment of Strength, Sensibility and Prehension (GRASSP) test and computerized measurement of the force generated by the isometric contraction of muscles (QMAD). Status:**
   GRASSP, a partnership between NACTN, the Canadian SCI Solutions Network and the European Clinical Trials Network, represents the kind of international collaboration envisioned when NACTN was initially organized – from www.sci-grassp.org -

Initiated by the North American Clinical Trials Network (NACTN) and the European Clinical Trials Network (EUCTN), a meeting was held on May 12 and 13, 2006 in Chicago (local organizer Drs. Zev Rymer and Lisa-Ann Wuermser, financially supported by the Reeve Foundation and Novartis) to discuss the measurement of hand impairment and function in patients suffering from tetraplegia.

Today, GRASSP is available for online purchase (http://www.sci-grassp.org/Purchase.html), has two publications and a third under review, has been presented at a series of meetings in the US and Canada and is now in the midst of a longitudinal multi-site study to define its responsiveness and establish an upper limb recovery profile (Appendices E, F). GRASSP is a tangible deliverable that benefits the international spinal cord community and although it pre-dated the emergence of NACTN's Neurological Outcomes Assessment Initiative (NOA), it can nevertheless be considered NOA's first success.

Quantitative Motor Assessment Device (QMAD) developed by Dr. Robert Grossman, Houston, TX, in collaboration with Kinea Design, Chicago, IL, is a reliable, standardized, and accurate way to evaluate the strength of voluntary muscle contractions during the course of neurological disease or injury. The utility of the device has been extended to capture biceps/triceps measurements and intrinsic hand strength and dexterity. The new device, called the Peg Restrained Intrinsic Muscle Evaluator (PRIME) was developed using funds supplementary to DOD support and is due for delivery in February/March 2011. A protocol to measure hand and arm muscle strength of neurologically impaired patients using PRIME was submitted to The Institute for Rehabilitation and Research (TIRR), Houston. Approval is pending. The protocol will also be submitted to The Methodist Hospital when a second device is available.

The primary objective of the study is to further develop, validate and bring into clinical practice this quantitative and sensitive method of measuring return of muscle function in patients who have sustained an acute spinal cord injury (SCI). The current standard of neurological assessment of SCI is the American Spinal Injury Association (ASIA) scale. The system was designed to gauge the extent of neurological damage. However, the grades (0-5) are non-linear with respect to the ranges of strength represented by each integer, and small changes in strength may not be detected using ASIA. This lack of sensitivity has undoubtedly contributed to the failure of potential beneficial SCI clinical trials of new therapy to provide unequivocal proof of efficacy despite promising laboratory studies. New molecular and surgical therapies are currently in clinical trials and in planning. The functional improvements that may be achieved by these therapies are likely to be modest, at least initially until dosage and timing are optimized. Without better outcome measurements, these clinical trials have a high probability of failure, setting back the cause of patients with SCI. Development of sensitive outcome measures is arguably the most important issue in SCI clinical trials research.

7. Begin development and validation of sensitive, reproducible outcome measures for use in clinical trials. Status:

NACTN's Neurological Outcomes Assessment Initiative (NOA) was launched in 2008 to develop, test, and validate sensitive and reliable outcome measures to detect small improvements in human clinical trials of therapies. Emphasis is on detecting incremental changes in patients such as improvements in neurological level and/or quantitative measures for ASIA A/B/C. Since its February 2009 organizing meeting at The Methodist Hospital, Houston, TX and the May 2009 (Louisville) and September 2009 (Dallas) NOA Task Force meetings, NACTN's NOA Committee has taken on responsibility for this task. To-date, three research awards have been issued by the Reeve Foundation to further NOA's goals:

1. Imperial College London, Peter Ellaway, PhD for Validation of the electrical perceptual threshold test as a quantitative assessment of cutaneous sensory function for spinal cord injury trials, (AppendixG)
2. University of British Columbia, Andrei Krassioukov, MD, PhD and University of Louisville, Louisville, KY, Susan Harkema, PhD for Natural Progression and recovery of cardiovascular parameters following traumatic spinal cord injury (Appendix H)
3. University of Louisville, Susan Harkema, PhD for Brain/Motor Control-EMG measures,

(Appendix I)

8. **Expanded NACTN contributes to ongoing Surgical Treatment of Acute Spinal Cord Injury Study (STASCIS). Status:**
Appendix J is a summary of the STASCIS study from a paper NACTN PI Michael Fehlings, MD, PhD will be submitting to the New England Journal of Medicine once all the co-authors have had a chance to review and comment. The central message conveyed in the abstract, that early decompression results in improved outcome, is very promising and exciting. The next steps for STASCIS are multi-fold in nature: a) examine the subgroups which most benefit from early surgical intervention; b) examine rates of secondary complication in detail; c) do a cost-effectiveness analysis; d) examine the roll-out of early surgical intervention as a best practice opportunity; e) examine opportunities for international collaboration; f) merge STASCIS and NACTN data to increase impact.

9. **Develop protocols for Phase II of anti-Nogo antibody treatment for SCI (ATI533). Status:**
ATI355 was temporally terminated in October of 2009 pending final screening of the last two patients enrolled to complete the total number to be enrolled. Both patients failed screening and enrollment was reopened until the end of February 2011. The Methodist Hospital has conducted preliminary over-the-phone screening of several potential SCI candidates. All have failed screening due to the strict criteria of the trial. Currently, 15 patients have been enrolled in Cohort 5 of the ATI533 phase I; a total of 16 is required. One of the 15 patients received 3 of the 6 planned intrathecal bolus injections. The patient had a SAE (pulmonary embolism), unrelated to ATI355 and unrelated to the injection mode of administration. That subject will be replaced. Therefore, two additional patients are needed. The Data Safety Monitoring Board will meet when the last patient of Cohort 5 has completed treatment. The Novartis management board will meet next month to review the current available A2102 Cohort 5 data and it is anticipated that the most likely the outcome will be an endorsement of an additional study in sensory-motor incomplete (AIS-C) subjects.

10. **Riluzole safety study. Status:**
See above at #1 for status of Riluzole protocol regulatory process.

The first therapy NACTN is testing is the neuroprotective agent, Riluzole, administered orally. The first patient was recruited on April 13, 2010 by the University of Maryland Shock Trauma; as of December 31, 2010, a total of 24 (out of 36) subjects has been recruited as follows:

- 8, University of Maryland
- 9, Thomas Jefferson University
- 4, University of Virginia
- 2, University of Texas Health Science Center, Houston
- 1, University of Louisville

To-date, no serious adverse effects have been reported due to the administration of Riluzole. Drs. Chow and Grossman and colleagues have developed an HPLC assay used to quantify Riluzole in small volumes of human plasma and CSF (manuscript in development). The Pharmacology Center has been monitoring study subjects for levels of the drug in their plasma; results are reported in Appendix K. Using non-DOD funding, Drs. Chow and Fehlings and colleagues have quantified Riluzole in rat plasma, brain, spinal cord and liver (manuscript in development).

On December 30, 2010, Charles H. Tator, MD, PhD, NACTN Principal Investigator (University of Toronto) and chair of the NACTN Treatment Strategy Selection Committee reported his Committee's initial deliberations and recommendations to Dr. Grossman for further consideration by NACTN's Executive Committee. A Phase III randomized prospective control trial of Riluzole, using an adaptive design model, is recommended (assuming the safety study is completed without adverse findings). Protocol development will commence in timely fashion.

11. **Stemnion study. Status:**
Efficacy of Amnion-Derived Multipotent Progenitor Cells (AMPCs) for Acute Treatment of Spinal Cord Injury -As of this writing, all the animal experiments for the proposed projects have been completed, including several additional directions approved as modifications to the contract and ACURO protocols.

Efficacy for AMPC transplantation and ACCS (amnion-derived cellular cytokine suspensions) administration via intrathecal catheter has been demonstrated and replicated, and final histology is currently in progress and will result in submission of a manuscript combining data from these multiple approaches. A final report on this contract is in preparation and will be submitted shortly.

12. Continued interactions with other clinical networks. Status:

- August 26 – 28, 2010 – NACTN investigators (Drs. Grossman, Tator, Fehlings, Harkema, Shaffrey, Harrop, Aarabi, Frankowski and Guest) attended a meeting hosted by the Reeve Foundation, the UK's International Spinal Research Trust and the Swiss Institute for Research in Paraplegia: *Spinal Cord Research: On the Way to Translation*. Drs. Grossman, Fehlings, Frankowski and Harkema met with investigators from EM-SCI to discuss the mechanics of data sharing between the two clinical networks
- November 3, 2010: Dr. Grossman, Susan Howley and Peter Wilderotter (President & CEO, Reeve Foundation) met with representatives of the National Institutes of Health, NIH attendees included: from NINDS - Dr. Story Landis, Director, Dr. Naomi Kleitman, SCI program officer and Marion Emr, Public Relations & Communications; from NICHD - Dr. Alan Guttmacher, Director. Dr, Mike Weinrich, and Dr. Ralph Nitkin. The meeting was an opportunity for Foundation to educate NIH representatives about its clinical research networks as prelude to a longer-term effort to identify ways NACTN and NRN can work synergistically with NINDS and NICHD. Dr. Landis observed that "there is support at NINDS for creating a mechanism to move therapies forward in SCI … we need to think about how the Reeve Foundation's goals can be accommodated within existing NINDS networks…" Perhaps the most important agreement that emerged from the meeting was acknowledgement at the NIH level that existing NIH-supported clinical trial networks do not serve SCI well because the requisite expertise is notably lacking in them. Dr. Grossman and Ms. Howley have a follow-up meeting scheduled with Dr. Landis and her team on February 25, 2011; it is hoped that Drs. Tator, Frankowski and Harkema will be able to join them at that time.
- December 3, 2010 – Dr. Grossman and Elizabeth Toups presented an abstract and poster on the Riluzole trial at the Annual Scientific Mission Connect Symposium on "Recent Advances in Basic and Clinical SCI Research and Therapy". Keynote speakers were Scott Whittemore, PhD, University of Louisville, and Aileen Anderson, PhD, University of California, Irvine. Dr. Whittemore's discussed *Are current experimental spinal cord injury models and treatments therapeutically relevant?* Dr. Anderson presented *Micromanaging the Microenvironment: The Impact of Inflammation on Stem Cell Repair Strategies for Spinal Cord Injury.*
- March 17 – 19, 2011 – will be the first joint meeting of the Reeve Foundation's two clinical research networks, NACTN and the NeuroRecovery Network. Principal Investigators and NRN Directors will spend two days in Toronto exploring opportunities for collaboration, including data sharing. With the field of spinal cord research moving inexorably toward clinical trials, this is a remarkable opportunity to bring together these unique networks in search of the most effective and rigorous ways to translate basic science findings to the clinic. The meeting will also include a discussion of NACTN-STASCIS data bases and NACTN PIs will attend an MRI workshop.

13. Administrative core activities. Status:

1. 2010 saw a paradigm shift in NACTN organizationally. In response to PLR recommendations, NACTN and the Reeve Foundation wrote and in March 2010, formally adopted a Governance Manual to inform network activities, deliberations and decision-making. A network-wide reorganization led to the distribution of responsibilities across committees. NACTN is now driven by an Executive Committee and Standing Committees include Publications, Data Management, NOA and Treatment Strategy Selection Committees. These committees meet by telephone conference monthly; minutes are taken and posted on the NACTN ftp site maintained by the Reeve Foundation.

    - Executive Committee (Chair, Robert G. Grossman, MD; Ralph Frankowski, PhD, Michael Fehlings, MD, PhD and Susan Harkema, PhD) – provides governance and addresses long-term issues critical to the goals and objectives of NACTN
    - The Publications Committee (Chair, James Harrop, MD) facilitates dissemination/ publication/presentation of NACTN data and insuring their integrity; this committee is also taking the lead identifying publication topics and developing the manuscripts

- NOA (Chair, Susan Harkema, PhD) guides NACTN's development, testing and validation of sensitive and reliable motor, autonomic, sensory, pain and quality of life outcome measures to detect incremental improvements in patients
- The Treatment Strategy Selection Committee (Chair, Charles Tator, MD, PhD) is charged with identification of potential new SCI therapeutics; a thorough review of all animal and preclinical data and then formulation of recommendations to NACTN's Executive Committee (Appendices L, M)

2. TMH-CC holds hour-long monthly teleconference calls for all NACTN coordinators (first 30 minutes, Q&A); Principal Investigators join for the final thirty minutes of the discussion.
3. The release of BAA-2010 funds enabled the Foundation to bring all NACTN centers, including the Data Management and Pharmacological sites, up to full funding for their 2010/2011 contracts.
4. The Reeve Foundation was given a no-cost extension on contract W81XWJ-07-1-0361 through December 31, 2011; this will enable us to complete the research tasks enumerated in our original proposal and in this award.
5. The Reeve Foundation requested and was granted budget modification requests on contract W81XWH-10-2-0042; these mods were necessitated by long lag times between original budget submissions and actual release of funding by DOD contracting and a request for a bridge loan that eventually materialized some 16 months later as a mod. The approved mods insure NACTN will continue to meet its research obligations envisioned in the BAA2010 proposal and, in a new initiative, establish a working relationship with the NeuroRecovery Network.

Key Research Accomplishments:

- Enrollment of 441 acutely injured patients into the NACTN data registry effective December 31, 2010 (#2)
- Initiation of NACTN's first clinical trial, the Phase I safety study of the neuroprotective drug Riluzole and enrollment of 24 of the total 36 subjects required (#10)
- Maturation of the Neurological Outcomes Assessment initiative and the award of NOA's first three research grants to develop and test three new instruments (#7)
- Affordable availability of GRASSP, a new clinical outcome assessment, for the entire spinal cord research community (#6)
- Submission of NACTN's first manuscript, Acute Complications after Spinal Cord Injury (Reportable Outcomes, below)
- Submission of NACTN's first chapter, Building a Clinical Trials Network for Spinal Cord Injury (Reportable Outcomes, below)
- Near-term publication of STASCIS findings and implications for future acute injury intervention (#8)
- Development of two manuscripts by NACTN's Pharmacology center (Reportable Outcomes, below)
- The re-organization of NACTN into a committee-based entity has established a hierarchy for decision-making, distributed responsibility for NACTN's operations and outcomes among the Principal Investigators, engaged many others on the NACTN team and insured continuity of leadership

Reportable Outcomes:

- Acute Complications after Spinal Cord Injury - A Multicenter Prospective Study of the Spectrum, Incidence and Severity: The North American Clinical Trials Network (NACTN) for Treatment of Spinal Cord Injury; Robert G. Grossman, Ralph F. Frankowski, Keith D. Burau, Elizabeth G. Toups, Michele M, Johnson, Michael G. Fehlings, Christopher I. Shaffrey Susan J. Harkema, Jonathan E. Hodes, Bizhan Aarabi, Michael K. Rosner, James D. Guest, James S. Harrop, submitted for publication (Appendix N)
- NACTN: Building a Clinical Trials Network for Spinal Cord Injury; Robert G. Grossman, Elizabeth Toups, Ralph Frankowski, Keith Burau, Susan Howley, for the NACTN Investigators in Essentials of Spinal Cord Injury; publisher Thieme; in press (Appendix O)
- Grossman, Robert G., Toups, Elizabeth G., for the NACTN investigators, A Phase I Clinical Trial of Riluzole, a Neuroprotective Drug, for Acute SCI, North American Clinical Trials Network for Treatment of Spinal Cord Injury for the NACTN Investigators; poster presentation, Mission Connect Symposium, December 3 2011 (Appendix P)

- Presentation, Reeve Foundation Spinal Cord Symposium, From Bench to Bedside, December 11, 2010 (Appendix Q)
- High Performance Liquid Chromatographic Assay for Riluzole in Rat Plasma, Brain, Spinal Cord and Liver; Yang Teng, Yongchao Wu, Michael G. Fehlings, Robert G. Grossman and Diana S-L. Chow; in development (Appendix R)
- High Performance Liquid Chromatographic Assay for Riluzole in Human Plasma and Cerebrospinal Fluid (CSF) Samples; Yang Teng, Elizabeth G. Toups, Robert G. Grossman and Diana S-L. Chow; in development (Appendix S)
- Presentation by Michael Fehlings, MD, PhD, at OTA Course, Extremity War Injuries VI: Data-Driven Progress in Combat Casualty Care," January 20, 2011; "Acute spinal cord injury: What is hot in clinical management and translational research?" Dr. Fehlings' presentation included data from the NACTN data registry, the phase I Riluzole safety study and STASCIS
- Kalsi-Ryan S, Beaton D, Curt A, Duff S, Popovic M, Rudhe C, Fehlings M, Verrier MC. Graded Redefined Assessment of Sensibility Strength and Prehension (GRASSP): Psychometric Development of an Upper Limb Impairment Measure for Individuals with Traumatic Tetraplegia; poster presentation: Sukhvinder Kalsi-Ryan (1st prize in student category). National SCI Conference, October 2010, Niagra Falls, Canada (Appendix T)

Conclusion:

At this writing, NACTN has addressed all the tasks enumerated in its original proposal; some have been completed successfully, others are works-in-progress that will continue to evolve over time. And because NACTN is a dynamic entity, unanticipated possibilities have presented themselves from time-to-time and the Foundation very much appreciates DOD's willingness to sanction an occasional new thrust or deviation from our original proposal.

Among recommendations by NACTN's Treatment Strategy Selection Committee is a Phase II/III study of Riluzole, pending successful conclusion of the ongoing Phase I. To insure the timely start of the larger trial, NACTN's Executive Committee will finalize the Phase II/III protocol now and initiate regulatory activities required for the study. It is hoped that by being anticipatory, lost time can be minimized and NACTN's momentum can be maintained.

The new DOD award insures two years of ongoing support that NACTN needs to continue to build its data registry, establish partnerships with other US and international clinical networks, conduct the Riluzole clinical trial, initiate others and facilitate NOA's development of outcome measures beyond those presently underway. Importantly, it will also enable the Foundation to expand NACTN beyond its current sites. As noted in the abstract (#14, above), Dr. Grossman has begun dialogue with Colonel Randall McCafferty about bringing San Antonio Military Medical Center into NACTN; a second new hospital will also be added as part of the scope of work of the new DOD contract.

We continue to be concerned about the sometimes unpredictable ebb and flow of funding and the future sustainability of that funding. To that end, we are exploring possible financial alliances with our Canadian colleagues and we have begun discussions with appropriate NIH representatives about ways to capitalize on NINDS' programs and NACTN's infrastructure, expertise and achievements. In the end, the ability of NACTN to fulfill its unique role in development, testing and delivery of new and effective treatments for spinal cord injury and its secondary dysfunctions depends on an uninterrupted revenue stream.

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Cahill et al. One Degree of Separation: Paralysis and Spinal Cord Injury in the United States. Released April 21, 2009

Cahill A, Fredine H, Zilberman L, Ibanez B, Fos M, Winneberger D. Overview of survey methodology: prevalence of paralysis including spinal cord injuries in the United States, 2008. Report from the University of New Mexico Center for Development and Disability. Released April 21, 2009

Grossman, Robert G., Frankowski, Ralph F., Burau, Keith D., Toups, Elizabeth G., Johnson, Michele M., Fehlings, Michael G., Shaffrey, Christopher J., Harkema, Susan J., Hodes, Jonathan E., Aarabi, Aarabi, Rosner, Michael K., Guest, James D., Harrop, James S., Acute Complications after Spinal Cord Injury - A Multicenter Prospective Study of the Spectrum, Incidence and Severity: The North American Clinical Trials Network (NACTN) for Treatment of Spinal Cord Injury; submitted for publication

Grossman, Robert G., Toups, Elizabeth, Frankowski, Ralph, Burau, Keith, Howley, Susan, NACTN: Building a Clinical Trials Network for Spinal Cord Injury; for the NACTN Investigators in Essentials of Spinal Cord Injury; publisher Thieme; in press

Kalsi-Ryan S, Beaton D, Duff S, Popovic M, Rudhe C, Curt A, Fehlings MG, Verrier MC. The Graded and Redefined Assessment of Sensibility Strength and Prehension (GRASSP) – Reliability and Validity. Submitted to Journal of Neurotrauma, Manuscript #: NEU-2010-1504

Kalsi-Ryan, Sukvinder, Curt, Armin, Fehlings, Michael G., and Verrier, Mary C., Assessment of the Hand in Tetraplegia Using the Graded Redefined Assessment of Strength, Sensibility and Prehension (GRASSP): Impairment Versus Function; Top Spinal Cord Inj Rehabil 2009;14(4):34–46

Kalsi-Ryan et al, Development of the Scoring Approach for the Graded Redefined Assessment of Strength Sensibility and Prehension (GRASSP); in development

Kalsi-Ryan et al, Relationships of Sensory and Motor Impairment to Prehension and Upper Limb Function in Tetraplegia; in development

Teng, Yang, Wu, Yongchao, Fehlings, Michael G., Grossman, Robert G., and Chow, Diana S-L, High Performance Liquid Chromatographic Assay for Riluzole in Rat Plasma, Brain, Spinal Cord and Liver; in development

Teng, Yang, Toups, Elizabeth, Grossman, Robert G., and Chow, Diana S-L High Performance Liquid Chromatographic Assay for Riluzole in Human Plasma and Cerebrospinal Fluid (CSF) Samples; in development

Spinal Cord Injury Information Network http://www.spinalcord.uab.edu. Facts and Figures at a Glance January, 2008

Appendices

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Quarterly Report Format

1. Award No. W81XWH-07-1-0361
2. Report Date January 24, 2011
3. Reporting period: August 16-December 31, 2010
4. Principal Investigator Dr. Robert Grossman
5. Telephone No.: 713-441-3810
6. Award Organization: Christopher Reeve Foundation
7. Project Title:North American Clinical Trials Network for Treatment of Spinal Cord Injury
8. Current staff, role and percent effort of each on project. **CONTINUED ON NEXT PAGE**

STAFF MEMBER	**Role**	**% EFFORT**
Robert Grossman MD	PI-Main	20
Susan Howley	Admin	21.65
Peter Wilderotter	Admin	1.5
Edward Jobst	Admin	4.25
Anne Homa	Admin	9
Bruce Morgan thru 12/31/08	Admin	10
Elizabeth Toups RN	Study Coordinator	40

9. Contract expenditures to date (as applicable):

COST ELEMENTS	**THIS QUARTER**	**CUMULATIVE**
Personnel		
Fringe Benefits		
Supplies		
Equipment		
Travel		
Other Direct Costs		
Subtotal		
Indirect Costs		
Fee		
Total		

10. Comments on administrative and logistical matters.

11. Use additional page(s), as necessary, to describe scientific progress for the quarter in terms of the tasks or objectives listed in the statement of work for this contract. Explain deviations where this isn't possible. Include data where possible.

12. Use additional page(s) to present a brief statement of plans or milestones for the next quarter.

8. Current staff, role and percent effort of each on project.

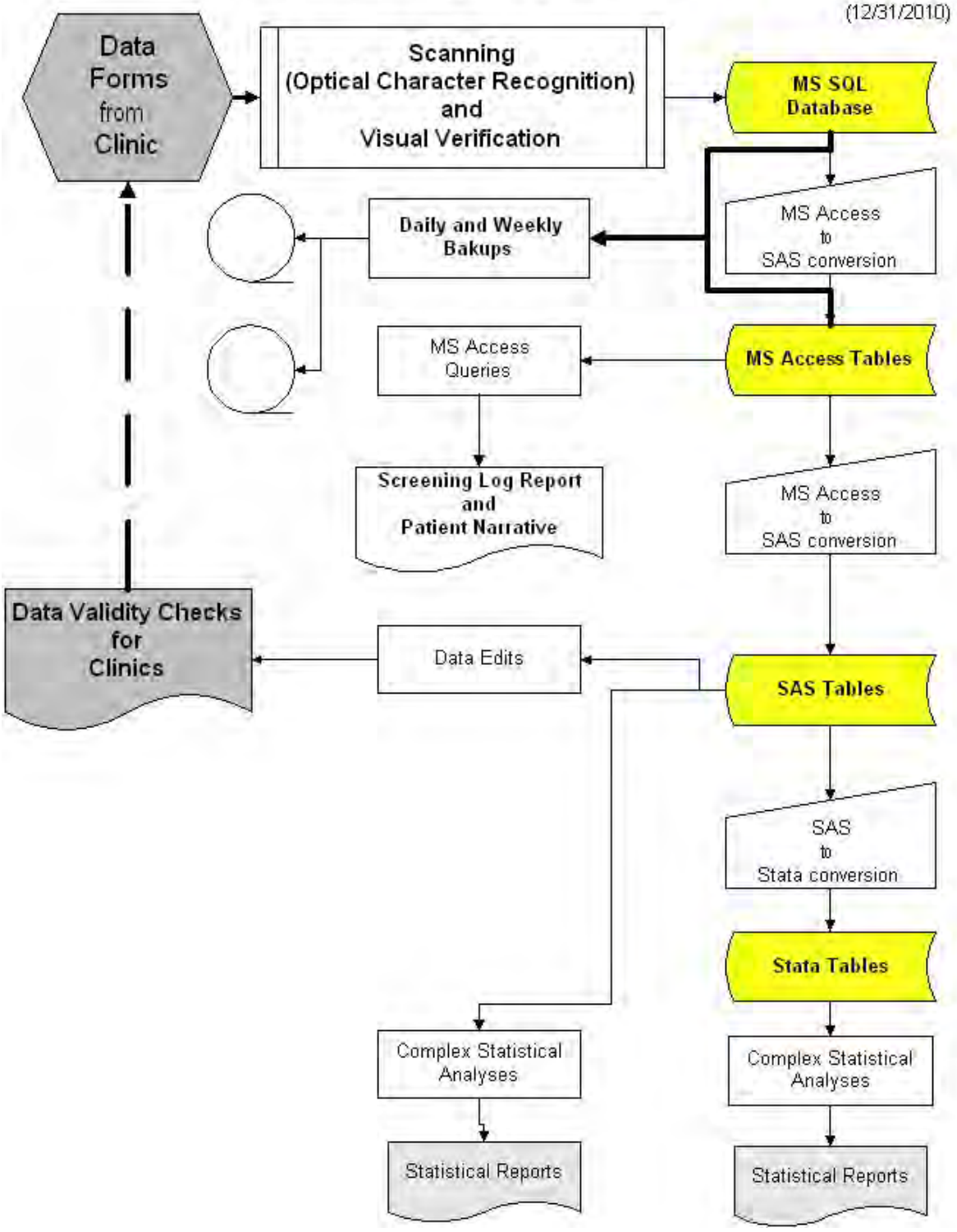
STAFF MEMBER	Role	% EFFORT
Michele Johnson, MD	PI	10
Michele Edelbrock	Study Coordinator	60
Michael Fehlings MD PhD	PI	15
Yuriy Petrenko MD	Study Coordinator	60
Christopher Shaffrey MD	PI	20
Daniel Chernavsky, MD	Study Coordinator	60
Susan Harkema, PhD, Jonathan Hodes, MD	Co-PIs	15
Elizabeth McDowell, RN	Study Coordinator	60
Bizhan Aarabi MD	PI	15
Heather Thomas, BS, CPhT	Study Coordinator	60
James Guest, MD	PI	20
Qing He, MD	Study Coordinator	100
Marina Dididze MD	Scientific Trials Coordinator	25
James Harrop, MD	PI	20
Deborah August MD	Study Coordinator	100
Michael K. Rosner MD	PI	20
Thomas Maryniak	Study Coordinator	75
TBD	Nurse Clinician	75
Ralph Frankowski PhD	PI	2.3
Keith Burau	Co PI	15
Hyvan Dang	Analyst	50
John DeLosReyes	Research Ass't	19
Nina Newton	Database Manager	50
Colleen Moore	Support Specialist	23
Tim Pierce	Grants	4
Michele Payton	Project Coordinator	4
Diana Chow	Co-PI	9.1
Yang Teng	Technician	50

Appendix A

NACTN Registry Data Flow

NACTN Registry Data Flow

(12/31/2010)



(12/31/2010)

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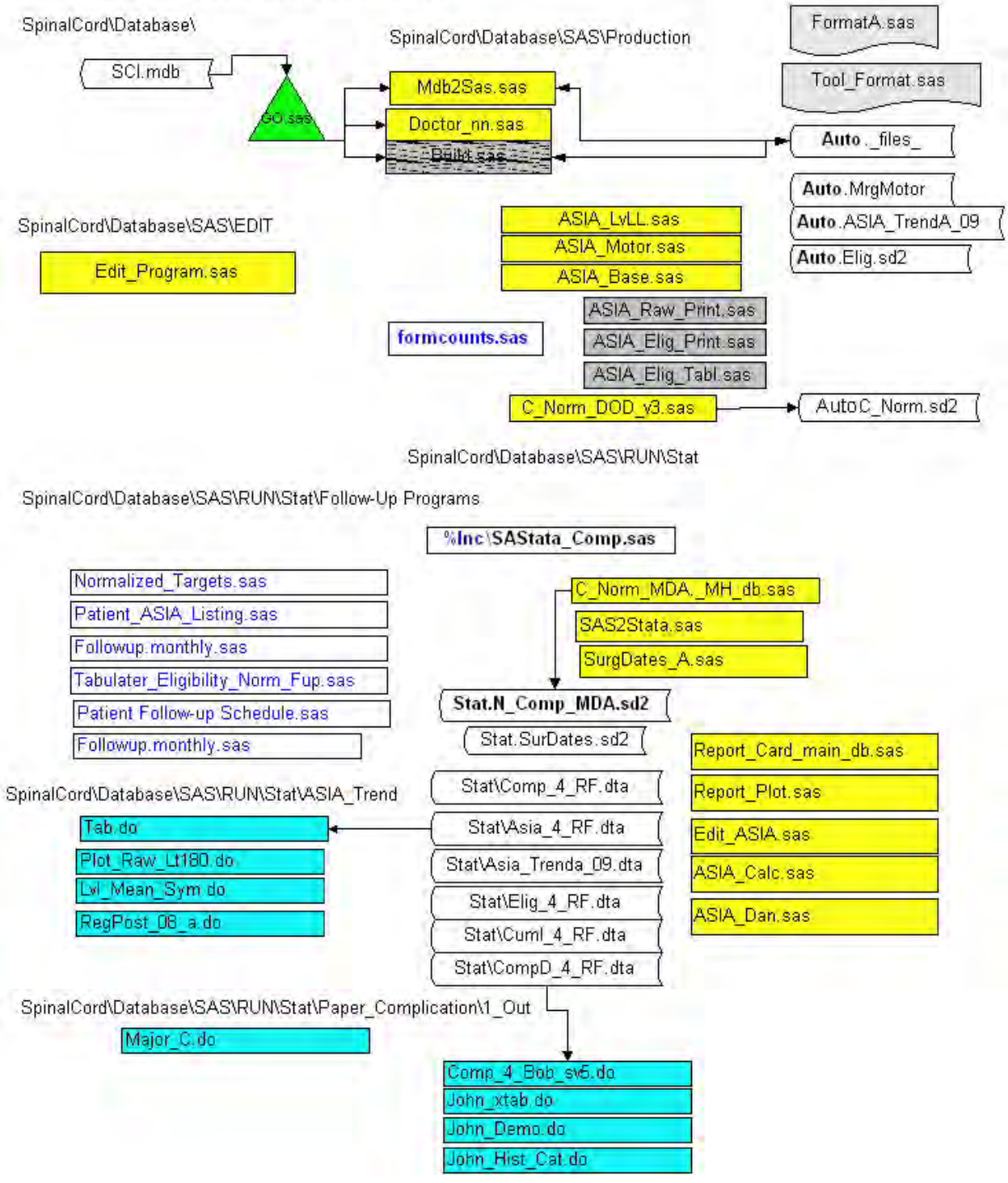

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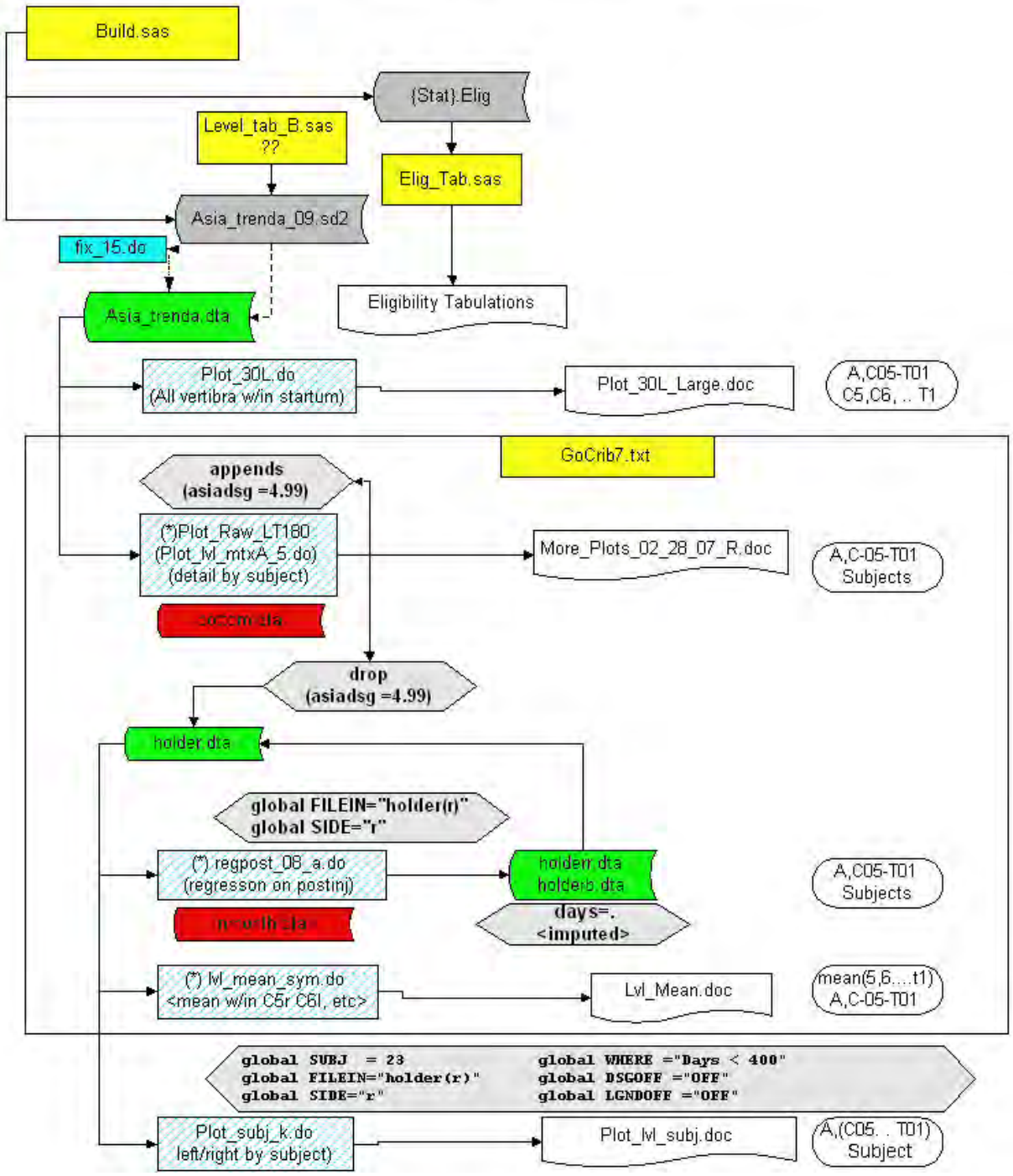

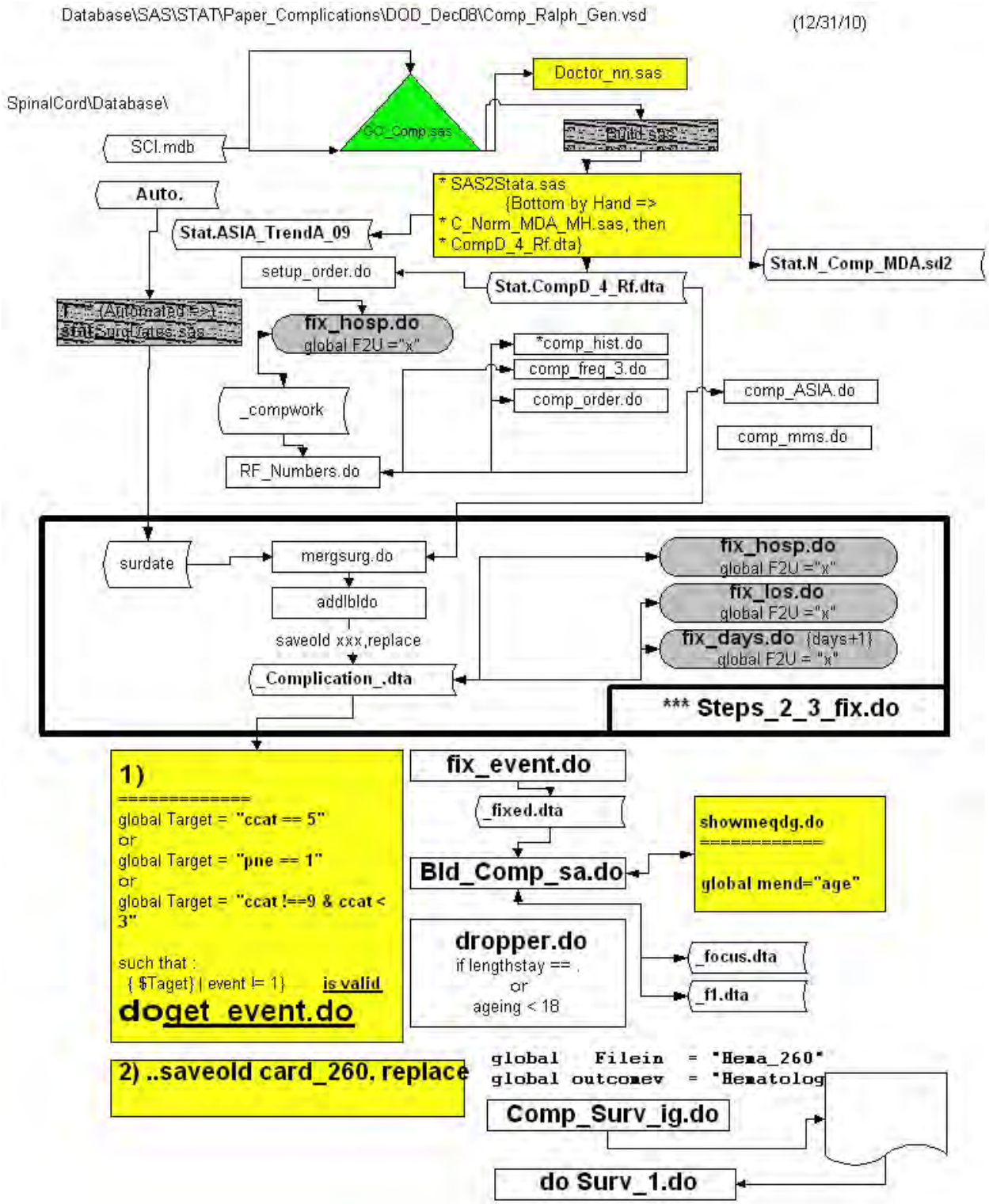
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Doctor_nn.sas
Bldtt.sas
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{Bottom by Hand =>
* C_Norm_MDA_MH.sas, then
* CompD_4_Rf.dta}
Auto.
Stat.ASIA_TrendA_09
Stat.N_Comp_MDA.sd2
setup_order.do
Stat.CompD_4_Rf.dta
fix_hosp.do
global F2U ="x"
*comp_hist.do
comp_freq_3.do
comp_order.do
comp_ASIA.do
comp_mms.do
_compwork
RF_Numbers.do
surdate
mergsurg.do
addlbldo
saveold xxx,replace
Complication.dta
fix_hosp.do
global F2U ="x"
fix_los.do
global F2U ="x"
fix_days.do {days+1}
global F2U = "x"
*** Steps_2_3_fix.do
1)
==========
global Target = "ccat == 5"
or
global Target = "pne == 1"
or
global Target = "ccat !==9 & ccat < 3"
such that :
{ $Taget} | event != 1}
is valid
doget_event.do
fix_event.do
_fixed.dta
Bld_Comp_sa.do
showmeqdg.do
==========
global mend="age"
dropper.do
if lengthstay == .
or
ageing < 18
_focus.dta
_f1.dta
2) ..saveold card_260, replace
global Filein = "Hema_260"
global outcomev = "Hematolog
Comp_Surv_ig.do
do Surv_1.do

Appendix B
North American Clinical Trials Network

SCI Data Registry Summary

06/25/2005 -12/31/2010

North American Clinical Trials Network

Table 1. Registry Screening and Enrollment

Status	Number	Percent Total
Screened	792	-
Enrolled	441	55.8%
In Database	407	92.3%
Pending	34	7.7%

North American Clinical Trials Network

Table 2. Patient Demographics

Characteristic	Number (N=407)	Percent
Gender		
Male	323	79.4
Female	84	20.6
Age[1] (yrs)		
< 20	29	7.1
20-65	329	80.8
>65	49	12.0
Race		
White	309	75.9
Other	98	24.1

[1]Median age at injury = 43 yrs of age

North American Clinical Trials Network

Table 3. Circumstances of Injury

Circumstance	Number (N=407)	Percent
Fall	146	35.9
MVA	125	30.7
Motorcycle	41	10.1
Recreation	47	11.5
Assault	25	6.1
Other	5	1.2
Unknown	18	4.4

North American Clinical Trials Network

Table 4. AIS Grade at Admission

AIS Grade	Number	Percent
A	142	34.9
B	54	13.3
C	49	12.0
D	101	24.8
E	36	8.8
Unknown	25	6.1
TOTAL	407	100.0

North American Clinical Trials Network

Table 5. Incidence of Complications

Complications	Number (N=407)	Percent
None	172	42.3
1	62	15.2
2	44	10.8
3	46	11.3
4+	83	20.4

North American Clinical Trials Network

Table 6. Complications by Type, Frequency, and Incidence

Complication Type	Frequency (N=1079(%)	Incidence Rate (%) (N=407 patients)
Pulmonary	285 (26.4)	36.4%
Infection	222 (20.6)	33.2%
Hematology	169 (15.7)	24.3%
Cardiac	138 (12.6)	23.3%
GI/GU	90 (8.3)	16.5%
Skin	89 (8.2)	16.5%
Neuropsychiatric	86 (8.0)	18.9%

North American Clinical Trials Network

Table 7. Injury Type and SCI Region

Characteristic	Number (N=407)	Percent
Injury Type		
Blunt	324	79.6
Crush	61	15.0
Penetrating	18	4.4
Other	4	1.0
Injury Region[1]		
Cervical	305	74.9
Thoracic	75	18.4
Lumbar/Sacral	24	5.9
SCIWORA	3	0.7

[1]Highest level reported when injury involved multiple levels

Table 8. Surgeries by AIS Grade

	Surgeries (N=407)				
AIS Grade	**Posterior**	**Anterior**	**Both**	**None**	**TOTAL**
A	67	19	44	12	**142**
B	24	12	12	12	**54**
C	22	11	10	6	**49**
D	32	41	16	12	**101**
E	14	2	1	19	**36**
Unknown	7	8	2	8	**25**
TOTAL	**166**	**93**	**85**	**63**	**407**

North American Clinical Trials Network

Table 9. Steroid Use by AIS Grade at Admission

	Steroids (N=407)		
AIS Grade	**Yes (%)**	**No (%)**	**N**
A	63	36	142
B	69	30	54
C	65	35	49
D	52	48	101
E	14	86	36
Unknown	80	20	25

North American Clinical Trials Network

Table 10. Hospital Stay and Discharge Status

Hospital Stay	Number (N=406)	Percent
< 8 days	106	26.1
8-14	115	28.3
15-21	69	17.0
> 21	116	28.6
Discharge Status	**Number (N=406)**	
Rehab Hospital	287	70.7
Home Care	84	21.6
Nursing Home	12	3.9
Long-Term Acute Care	9	
In-Hospital Deaths	14	

North American Clinical Trials Network

Table 11. ASIA Grades – Admission vs. Discharge

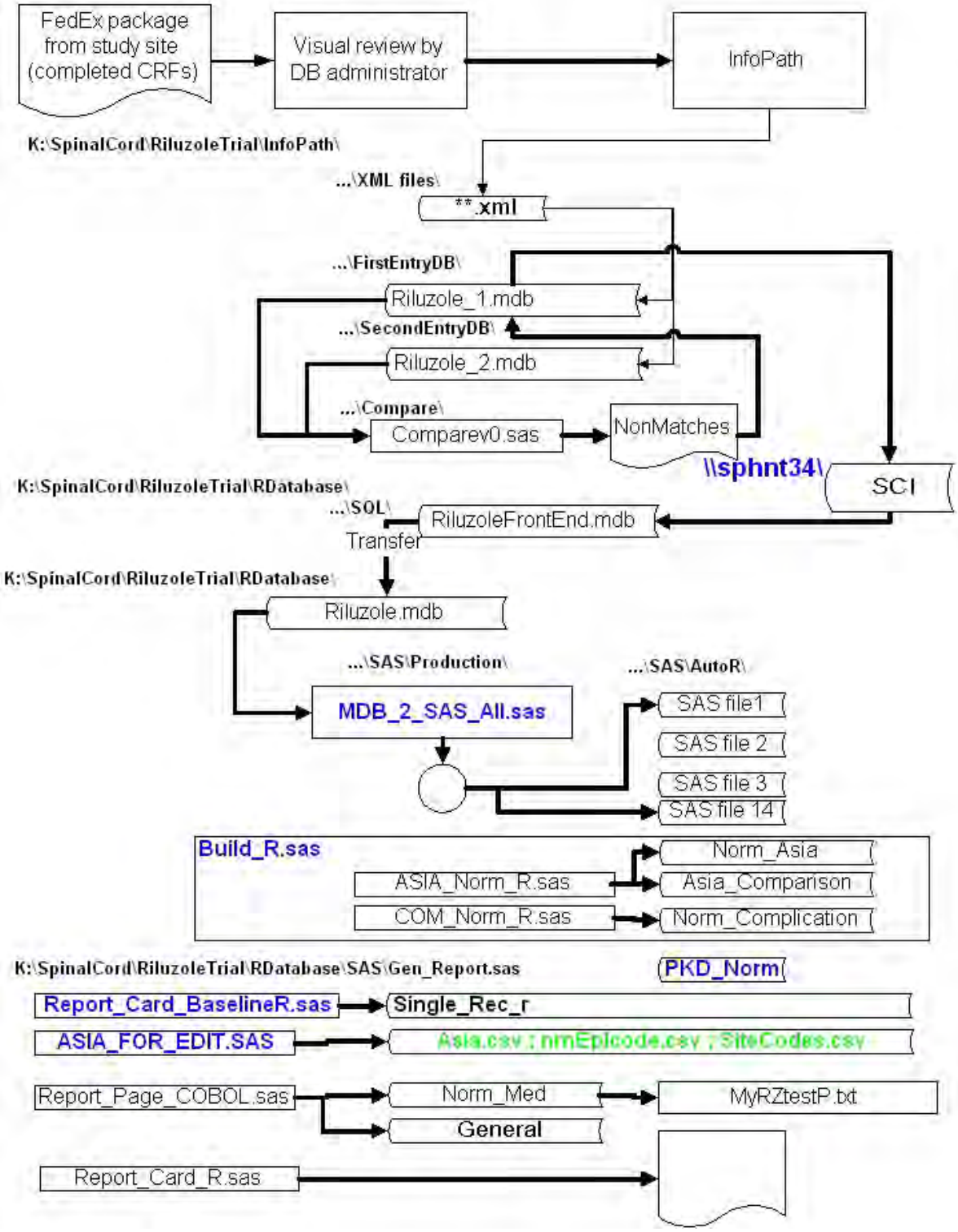
	AIS Discharge					
AIS Admit	**A**	**B**	**C**	**D**	**E**	**Total**
A	115	10	5	0	0	**130**
B	7	30	13	2	0	**52**
C	1	1	30	16	0	**48**
D	0	0	4	89	8	**101**
E	0	0	0	0	34	**34**
Total	**123**	**41**	**52**	**107**	**42**	**365**

Appendic_C

Data Flow for NACTN Riluzole Safety Trial

Data Flow for NACTN Riluzole Safety Trial

(12/31/2010)



Appendix D
Baseline Characteristics of Riluzole Patients
04/12/2010 – 12/31/2010

Table 1. Therapeutic Time Windows

Time	**minimum**	**25th percentile**	**median**	**75th percentile**	**maximum**
Injury to Admission	0.7 hrs	1.6 hrs	3.0 hrs	4.2 hrs	7.0 hrs
Injury to Treatment	5.5 hrs	6.9 hrs	8.5 hrs	10.7 hrs	12.0 hrs

Table 2. Demographics

Characteristic	**N = 20 (percent)**
Gender	
male	20 (100%)
Age	
Mean Age	37.4 years
25th percentile	20.5 years
Median Age	26.0 years
75th Percentile	55.0 years
Ethnicity	
White	12 (60%)
African-American	5 (25%)
Asian/Other	3 (15%)

Table 3. Circumstances of Injury

Circumstances	**N = 20 (percent)**
Motor Vehicle	8 (40%)
Fall	5 (25%)
Assault	2 (10%)
Diving	3 (15%)
Motor Cycle	1 (5%)
Bicycle	1 (5%)

Table 4. AIS Grade and SCI Severity

AIS Grade	**N = 20 (percent)**
AIS A	9 (45%)
AIS B	6 (30%)
AIS C	5 (25%)

SCI Severity	**N = 20 (percent)**
Tetra Incomplete	10 (50%)
Tetra Complete	3 (15%)
Para Incomplete	1 (5%)
Para Complete	6 (30%)

Table 5. Surgeries and Steroid Use

Surgery	**N=20 (percent)**
None	2 (10%)
Anterior	2 (10%)
Posterior	7 (35%)
Anterior + Posterior	7 (35%)
Pending DMC Submission	2 (10%)

Steroid Use	**N=20 (percent)**
No	13 (65%)
Yes	5 (25%)
Pending DMC Submission	2 (10%)

Title: Graded Redefined Assessment of Sensibility Strength and Prehension (GRASSP): Psychometric Development of an Upper Limb Impairment Measure for Individuals with Traumatic Tetraplegia

Authors and Investigators: Sukhvinder Kalsi-Ryan, MSc [1, 3, 5, 6, 11]; Dorcas Beaton, PhD [2, 3, 5, 8]; Armin Curt, MD [10, 11]; Susan Duff, PhD [9, 11]; Milos Popovic, PhD [3, 4, 5, 7]; Claudia Rudhe, MScOT [10, 11]; Michael G. Fehlings, MD [5, 6, 11]; Mary C.Verrier, MHSc [1, 3, 5, 7, 11]

Institutions: [1]Dept. of Physical Therapy, [2]Dept of Occupational Therapy [3]Graduate Dept. of Rehabilitation Science, [4]IBBME, [5]University of Toronto; [6]Krembil Neuroscience Centre, University Health Network; [7]Toronto Rehabilitation Institute; [8]St. Michael's Hospital, Mobility Evaluation and Clinical Research Unit, Toronto, Ontario; [9]Thomas Jefferson University, Philadelphia, Pennsylvania; [10]Balgrist University Hospital, Zurich; [11]International GRASSP Research and Design Team

Background and Significance: Upper limb function being of great importance to individuals with tetraplegia has lead to concentrated efforts in the field of neural repair and neuro-rehabilitation research. As new methods are established to enhance upper limb function by enhancing neurological recovery; a specific and sensitive measure of upper limb impairment was required to establish efficacy of interventions. Therefore, the Graded Redefined Assessment of Strength Sensibility and Prehension (GRASSP) was developed to fill this void.

Objectives: The overall objective of this work was to design and develop a sensitive, clinical upper limb impairment measure specific to tetraplegia; which could be implemented across the continuum of care and to the entire tetraplegic population. The four objectives addressed in this study are defined below:
1) To finalize and confirm the elements of GRASSP Version 1.0.
2) To define the scoring approach for the GRASSP Version 1.0.
3) To establish inter-rater and test retest reliability, construct/concurrent validity.
4) To determine relationships of domains within GRASSP and GRASSP domains to upper limb function.

Methods: A cross sectional multi-centre trial with repeated administration of the GRASSP by multiple examiners and single administration of validation measures was conducted in 4 centres in North America and 3 sites in Europe. The International Standards of Neurological Classification for Spinal Cord Injury (ISCSCI), Spinal Cord Independence Measure II (SCIM), Capabilities of Upper Extremity Function (CUE) and repeated GRASSP (x3) were administered on a sample of 72 individuals with chronic tetraplegia. **Analysis:** Regression analysis was applied to all items of all domains in GRASSP to guide the item reduction process. Guttman scaling to develop the scoring system; intraclass correlation coefficients to establish reliability, Pearson correlation coefficients to establish validity with SCIM and CUE were used. Structural equation modeling was used to establish relationships between GRASSP domains and function.

Results: Content and the manual for the GRASSP Version 1.0 was finalized. GRASSP subtests defined individual impairment and demonstrated cumulative predictive patterning 80% of the time, therefore, an individual is defined by 5 subtest scores for each extremity which can be plotted on a radar diagram. Inter-rater and test retest reliability ranged from 0.84-0.99. Construct validity (sensitivity) was confirmed by decreased level of agreement (kappa statistic 0.412-0.511) among GRASSP and ISCSCI sensory and motor items, supporting that the GRASSP testing provides greater precision of the upper limb status. GRASSP subtests demonstrated concurrence with the SCIM and CUE. Impairment showed the strongest concurrence with self-perception of function (0.57-0.83, $p<0.0001$). Quantifying impairment showed that sensation and strength both have direct and indirect influences on upper limb function.

Conclusion: The GRASSP is reliable, valid and is sensitive in defining upper limb impairment and function. GRASSP Version 1.0 is now available and should be used to track neurological functional changes longitudinally.

Funding: Christopher and Dana Reeve Foundation, Rick Hansen Foundation, Ontario Neurotrauma Foundation, Toronto Rehabilitation Institute Scholarship Fund and Physiotherapy Foundation of Canada

Website:
www.sci-grassp.org

Manuscripts:
1. Kalsi-Ryan S, Curt A, Fehlings, MG and Verrier MC. Assessment of the Hand in Tetraplegia Using the Graded Redefined Assessment of Strength Sensibility and Prehension (GRASSP): Impairment versus Function. Top Spinal Cord Inj Rehabil 2009;14(4):34-46.
This manuscript presents the analysis and findings to report Objective 1 - To finalize and confirm the elements of GRASSP Version 1.0. ATTACHED

2. Development of the Scoring Approach for the Graded Redefined Assessment of Strength Sensibility and Prehension (GRASSP).
This manuscript is currently being formatted for submission to Archives of Physical Medicine and Rehabilitation and presents the analysis and findings to report Objective 2 - To define the scoring approach for the GRASSP Version 1.0.

3. Kalsi-Ryan S, Beaton D, Duff S, Popovic M, Rudhe C, Curt A, Fehlings MG, Verrier MC. The Graded and Redefined Assessment of Sensibility Strength and Prehension (GRASSP) – Reliability and Validity. Submitted to Journal of Neurotrauma, Manuscript #: NEU-2010-1504
This manuscript is submitted and under review and presents the analysis and findings to report Objective 3 – To establish inter-rater and test retest reliability, construct/concurrent validity.

4. Relationships of Sensory and Motor Impairment to Prehension and Upper Limb Function in Tetraplegia.
This manuscript is currently being formatted for submission to Neurorehabilitation and Neural Repair and presents the analysis and findings to report Objective 4 - To determine relationships of domains within GRASSP and GRASSP domains to upper limb function.

Presentations/Posters:

1. Kalsi-Ryan S, Duff S, Rudhe C, Wuermser L, Curt A, Fehlings MG, Verrier MC. The GRASSP Protocol – The Value of Spinal Cord Injury (SCI) Research Networks to the Development of Outcome Measures. 2007. *Journal of Spinal Cord Medicine* Vol. 30(2): pp168
Poster presented by: Sukhvinder Kalsi-Ryan
Meeting: Annual American Spinal Injury Association Meeting, Tampa FL, June 2007.

2. Kalsi-Ryan S, Duff S, Rudhe C, Wuermser L, Curt A, Fehlings MG, Verrier MC. The Graded and Redefined Assessment of Sensibility Strength and Prehension (GRASSP Protocol) – Development and Validation of a Hand Function Measure for the Tetraplegic Population.
Podium Presentation: Sukhvinder Kalsi-Ryan
Meeting: International Meeting of Upper Limb Management in Tetraplegia, Philadelphia PA, Sept 2007

3. Kalsi-Ryan S, Duff S, Rudhe C, Curt A, Fehlings MG, Verrier MC. Reliability and Validity of the Graded and Redefined Assessment of Sensibility Strength and Prehension (GRASSP). 2008. *Journal of Spinal Cord Medicine* Vol. 31
Podium presentation: Sukhvinder Kalsi-Ryan
Meeting: Annual American Spinal Injury Association Meeting, San Diego, June 2008

4. Kalsi-Ryan S, Duff S, Rudhe C, Curt A, Fehlings MG, Verrier MC. A Reliable and Valid Measurement Approach for Hand Impairment In Tetraplegia: The Graded and Redefined Assessment of Sensibility Strength and Prehension (GRASSP).
Poster Presentation: Sukhvinder Kalsi-Ryan
Meeting: National SCI Conference, November 2008. Toronto, Canada

5. Sylvie Nadeau, Robert Forget, Dany Gagnon, and Sukhvinder Kalsi-Ryan. Clinical Workshop Presentation: Sensory-motor assessment of individuals with spinal cord injury.
Meeting: National SCI Conference, November 2008. Toronto, Canada

6. Kalsi-Ryan s, Beaton D, Curt A, Duff S, Popovic M, Rudhe C, Fehlings M, Verrier MC. Graded Redefined Assessment of Sensibility Strength and Prehension (GRASSP): Psychometric Development of an Upper Limb Impairment Measure for Individuals with Traumatic Tetraplegia. ATTACHED
Poster Presentation: Sukhvinder Kalsi-Ryan (1st prize in student category)
Meeting: National SCI Conference, October 2010, Niagra Falls, Canada

7. Kalsi-Ryan S, Kapadia N, Holmes J, Verrier M. Upper Limb Sensorimotor and Functional Assessment for Individuals with Tetraplegia - Graded Redefined Strength, Sensibility and Prehension (GRASSP).
Clinical Workshop
Meeting: National SCI Conference, October 2010. Niagra Falls, Canada

Ongoing and Current Work:
A longitudinal study engaging multiple sites in Canada and Europe is underway for which the objectives are to define responsiveness of the GRASSP, establish minimally clinical important differences and a recovery profile for the upper limb. This current study is being funded by the Ontario Neurotrauma Foundation and the SCI Solutions Network.

Assessment of the Hand in Tetraplegia Using the Graded Redefined Assessment of Strength, Sensibility and Prehension (GRASSP): Impairment Versus Function

Sukhvinder Kalsi-Ryan, Armin Curt, Michael G. Fehlings, and Mary C. Verrier

Objective: To refine the Graded and Redefined Assessment of Strength, Sensibility and Prehension (GRASSP) as a measure of upper limb impairment following cervical SCI. **Method:** A cross-sectional study assessed a cohort of neurologically stable patients with tetraplegia using a preliminary version of the GRASSP. Regression analysis was performed to determine the association between subcomponents of the GRASSP (impairment) and measures of function. The GRASSP was modified based on results. **Results:** Eliminated static two-point discrimination, tone, and one muscle. **Conclusion:** The GRASSP Version I consists of Semmes Weinstein monofilaments, manual muscle testing, and qualitative and quantitative prehension testing. **Key words:** *assessment, measurement, sensory motor impairment, tetraplegia, upper limb*

The ability to use the upper limbs is of central importance for individuals with tetraplegia as upper limb function determines overall function for these individuals. Not only do they use their hands to perform normal functions, but they also use their upper limb function as a substitute for other functions that are no longer possible. The upper limbs of an individual with tetraplegia are integral for

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activities such as locomotion, bowel and bladder care, recreation, and employment. Individuals with tetraplegia have identified upper limb function as one of the most significant factors contributing to quality of life.[1,2] Therefore, the more extensive upper limb recovery is following tetraplegia, the more functional an individual should be. In essence, upper limb function can equate to independence and global function for someone with tetraplegia.

Many investigators have studied ways to enhance upper limb movement (e.g., functional electrical stimulation and tendon transfer)[3,4] and have subsequently assessed outcomes by measuring elements that were thought to be impacted by the interventions as opposed to changes in impairment or global function of the upper limb. Existing approaches to assessment have measured targeted parameters such as force, magnitude, and duration of grasp.[5–7] Values for grasp parameters, however, do not necessarily reflect subtle neurological change[8,9]—change that may facilitate a more optimal movement pattern and improved hand function. Furthermore, subtle neurological change may be the only initial positive result observed with neuroprotective and neuroregenerative therapies in humans.[10,11] To assess efficacy, one needs to determine the degree of change required to optimize function. Therefore, a comprehensive and sensitive measure of upper limb impairment/function is needed to document change post injury. Such a measure, which would depend on multiple factors such as the interplay between the sensory and motor domains of movement, is essential for future interventions intended to improve neurological recovery after spinal cord injury (SCI).

Assessment of upper limb recovery after tetraplegia will also be important to future pharmaceutical trials. Recently, due to the difficulty with assessment of the thoracic area, increased consideration is being given to enrolling subjects with cervical SCI in trials studying biological and pharmacological agents.[12,13] It is hypothesized that neurological improvement in the cervical spinal cord is more likely to be reflected and detected as a change in upper limb function as compared to thoracic changes, which are difficult to assess. Furthermore, enrolling individuals with tetraplegia increases the number of potential subjects for studies, as almost two thirds of SCIs are cervical.[14] Increasing survival rates for cervical SCI have also driven interest in the development of a sensitive outcome measure for upper limb impairment. Researchers and experts have criticized prior trials[15] that used the International Standards for Neurological Classification of SCI, including the American Spinal Injury Association Impairment Scale (AIS),[16] as a primary outcome measure. The AIS was created and intended to be used as a clinical measure to classify injury severity, not as an outcome measure for efficacy in clinical trials. Nonetheless, the AIS has been utilized in many studies, and progress from human clinical trials has been hampered by the absence of a sensitive test for upper limb impairment, specifically the hand.

The first concentrated attempts by Sollerman and Ejeskar to measure hand function in the tetraplegic population met with limited success.[17] The Sollerman Hand Function Test was designed based on the conceptualization of normal hand function and did not adequately account for the impact of varying degrees and levels of cervical cord damage on hand impairment. Another outcome measure, the Danish Tetraplegia Hand Mea-

sure,[18] was designed to measure the ability to complete functional tasks performed using a passive tenodesis grasp. Approaches to the use of tenodesis grasp are not universal, and the lack of specific protocols limits the utility of the test to certain parts of the world and a selective subgroup of individuals. The Jebsen-Taylor Hand Function Test[19] is commonly used in SCI but was neither validated nor designed specifically for neurological populations. The Rehabilitation Engineering Laboratory Hand Function[20] and Grasp and Release[21] tests are specifically designed to assess the effects of functional electrical stimulation and neuroprosthetic interventions and have not been adopted universally. The Van Lieshout Test[22] was designed to assess upper limb capacity in tetraplegia and tests performance on tasks related to daily living. It has inter- and intrarater reliability of 0.98 and 0.99 (n = 12), respectively, and moderate concurrent validity with the Grasp and Release Test. Although useful during the subacute phase of recovery, the aforementioned functional tests are not feasible for use in the acute phase where new biological and pharmacologic interventions are targeted. Assessment of subtle change is paramount. Improved measures of upper limb impairment and function are required to determine efficacy in clinical trials and will need to be incorporated into Phase 2 and Phase 3 trials.[23]

Development of the GRASSP

It became clear to the pharmaceutical industry and scientists in the field that approaches to measure and determine the efficacy of emerging therapies were lagging and an outcome measure was needed that was both sensitive and responsive to change—one that could be used to track natural recovery and the response of individuals receiving treatment. These issues served as the rationale for the development of the Graded and Redefined Assessment of Strength, Sensibility and Prehension (GRASSP).

In May 2006, the North American Clinical Trials Network held a workshop in Chicago, Illinois, funded by the Christopher and Dana Reeve Foundation and Novartis International AG, Basel, Switzerland. The focus of the workshop was the discussion of the measurement of hand impairment and function in patients suffering from cervical SCI. Clinical specialists in hand measurement, rehabilitation practitioners, and SCI researchers with expertise in upper limb neurophysiology, engineering, and computer technology discussed the development of a comprehensive protocol to assess upper limb impairment and recovery post cervical SCI. At the conclusion of the meeting, a task force was formed to develop a new clinical protocol to assess upper limb (hand) impairment by modifying existing tools and introducing new measures intended to quantify changes in hand impairment starting immediately post injury. This led to the development of the alpha version of the GRASSP.

Theoretical Framework

The overall objective for the assembly of the GRASSP was the development of a clinical research measure that could (a) capture information on upper limb impairment for the cervical (C0-T1) SCI population, including data on integrated sensory and motor impairment; (b) discriminate according to the level of lesion; and (c) capture changes in hand impairment throughout the recovery

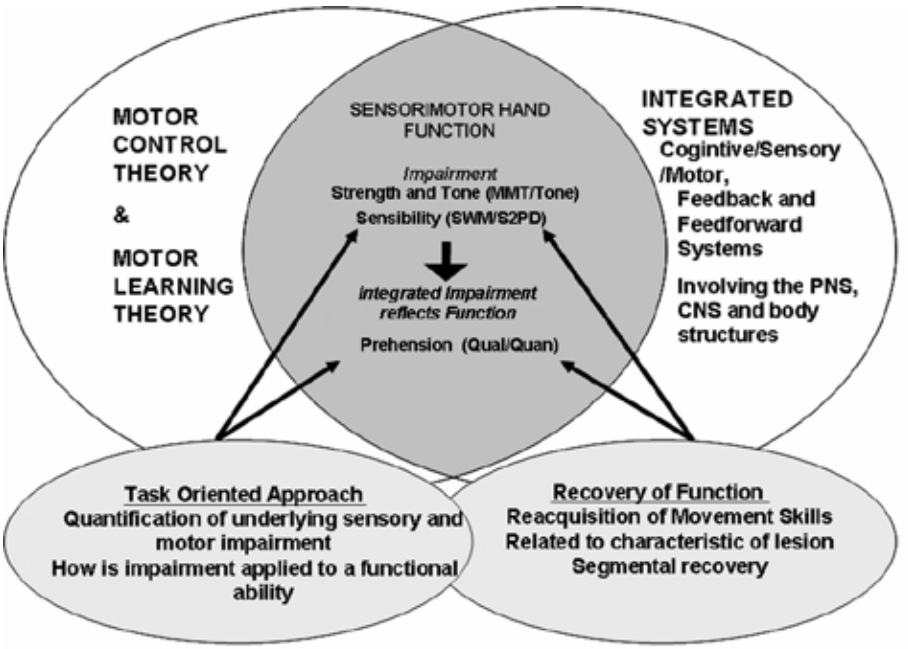

Figure 1. Theoretical framework for the development of the GRASSP.

phase. Sensorimotor function was defined as the major construct for the GRASSP, and a theoretical framework (**Figure 1**) was designed to guide development of the measure. The framework incorporated the concepts of motor control and motor learning theory,[24] which involve the interactions of the function (task), the individual, and the environment.[25] Task performance, which is dependent on integrated systems of sensation, motor, and cognition, was also incorporated. An integrated component was incorporated to assess how sensory and motor impairments contribute to an integrated function; this issue becomes increasingly important during the recovery process. When scoring is directed toward the quality and performance of movement (noting how the grasp is produced) more so than the ability alone (task performed or not), the results indicate which neurological elements are intact.

The initial GRASSP combined the preexisting Link Hand Function Test (LiHFT)[26] and the Tetraplegia Hand Measure (THM)[27] and incorporated three domains: strength and tone, sensibility (sensation), and prehension (integrated). The three domains provide the basis for the name of the measure, the Graded and Redefined Assessment of Strength, Sensibility and Prehension (GRASSP). The inclusion of multiple domains ensures comprehensiveness of assessment. Each domain can be tested individually or in conjunction with another. Prior to 3 to 4 weeks post injury,

it is recommended that a partial GRASSP be administered that consists of sensibility, strength and tone, and qualitative prehension only, because it is unlikely that the patient will tolerate enough sitting for the quantitative grasp portion of the test. However, if an individual is able to tolerate 45 minutes of sitting, a full GRASSP should be administered.

Phase I

Clinimetric development of the LiHFT and THM occurred during individual test development by Link[26] and Kalsi-Ryan et al,[27] respectively. Clinimetric development refers to the process of evaluating the clinical measurement properties of an assessment, such as feasibility, face, and content validity. The components of each test that met the criteria of the framework (see **Figure 1**) were selected and combined to create the GRASSP. Components adapted from the LiHFT included five prehension tasks. In a similar fashion, the sensory module, part of the motor testing, and the scoring scale from the prehension tasks (combined with the LiHFT scoring scale) were adapted from the THM. All components included in the GRASSP are presented in **Table 1**. The sensibility domain was assessed using Semmes Weinstein monofilaments (SWM) for light touch and static two-point discrimination (S2PD) for functional sensation. The strength/tone domain was assessed using manual muscle testing (MMT) for strength[28,29] and the Modified Ashworth Scale for tone.[30] Both descriptive and performance-based prehension tests were incorporated to address the prehension domain. The descriptive prehension test evaluates whether the thumb and digits can assume three specific grasps or can perform any active movement at all. The performance-based prehension test is a modified version of the Sollerman Hand Function Test.[17] The Sollerman was modified by Link and Kalsi-Ryan et al during the development of the LiFHT and the THM. The prehension domain in the GRASSP retains the Sollerman concept of evaluating specific activity of daily living (ADL) tasks performed with specific grasps for evaluation. Details of the modifications made to the Sollerman Hand Function Test are available in the **Appendix**. **Table 1** provides a summary of how the GRASSP is administered.

Phase II

Following initial development of the GRASSP, a cross-sectional study was used to determine which preliminary components should be included in the final GRASSP (GRASSP Version I). Seven centers collected data: Rehabilitation Institute of Chicago, Chicago, Illinois; Toronto Rehabilitation Institute, Toronto, Ontario; Vancouver Coastal Health, Vancouver, British Columbia; Thomas Jefferson University, Philadelphia, Pennsylvania; Balgrist University Hospital, Switzerland; Krakenhaus Hohe Worte, Germany; Traumacenter Murnau, Germany. Descriptive details for the study cohort (n = 72) are provided in **Table 2**. Additional details of Phase II are outlined below.

Initial Evaluation and Refinement of the GRASSP

Regression analysis was conducted to determine which tests to include in the final GRASSP and to create a clinical index and/or global score; however, sample size was not sufficient to perform the latter analysis. The GRASSP, Spinal Cord Independence Measure (SCIM),[31] Capabilities of Upper

Table 1. Components of the GRASSP and methods of administration

Components of the GRASSP			Method for administration	
Test	**Details**	**Rationale**	**Position time required**	**How to test**
Sensibility domain: test sites selected by dermatome				
Light touch/SWM[33]	6 palmar/dorsal test sites	Inter/intra-reliability = 0.965	Supine/ sitting, 10 min	-Apply monofilaments to all test locations -Summate the score for each hand separately
Static 2 Point Disc[33]	3 palmar test sites	Inter/intra-reliability = 0.989	Supine/ sitting, 5 min	-Apply stimulus to all test locations -Summate score for each hand
Strength and tone domain: muscle selection based on myotomes				
Strength[34]	MMT-4 arm & 7 hand muscles	Inter-reliability = 0.880	Supine/ sitting, 10 min	-Assess each muscle and grade -Summate all scores for each hand
Tone[30]	Modified Ashworth for hand & arm	Inter-reliability = 0.750	Supine/ sitting, 5 min	-Assess elbow and hand for flexor/ extensor tone
Prehension domain: segmental influence movement pattern				
Qualitative (descriptive)	3 grasps rated on scale of 0–4		Supine/ sitting, 5 min	-Have subject perform grasps and rate
Quantitative (performance)[17]	5 grasps/6 tasks rated on scale of 0–5	Adapted from Sollerman, inter-reliability = 0.980	Sitting, 15 min	-Set patient up in sitting at table and have patient perform all 6 tasks for each hand separately

Note: In Version I of the GRASSP, MMT, SWM palmar, SWM dorsal, Qualtiative Prehension, and Quantitative Grasp totals are then plotted on a polar diagram for interpretation. GRASSP = Graded Redefined Assessment of Strength, Sensibility and Prehension; MMT = manual muscle testing; SWM = Semmes Weinstein monofilaments.

Table 2. Study cohort

Study site	*N*	Description of sample
Toronto Rehabilitation Institute, Canada	15	C6-C7 AIS motor level, 52.5% C4-C6 AIS sensory level, 66.0% *AIS grades* A, 38.8% B, 25.2% C, 16.6% D, 19.4%
Vancouver Coastal Health Canada	10	
Rehabilitation Institute of Chicago, USA	10	
Thomas Jefferson University, USA	10	
Balgrist University Hospital, Switzerland	9	
Krakenhaus Hohe Worte, Germany	8	
Traumacenter Murnau, Germany	10	
Total	**72**	

Extremity Questionnaire (CUE),[32] and International Standards for Neurological Classification of SCI (ISNCSCI) were administered to all study participants. General linear modeling was then used to establish the strength of the association between the components of the GRASSP (impairment) and function as defined by the SCIM (a measure of global function), the SCIM self-care subscore (a measure of upper limb function), and performance-based prehension tasks from the GRASSP (a measure of hand function). Individual subscores for each test within the GRASSP were calculated. SWM scores were separated into palmar and dorsal scores. GRASSP subscores were then compared to functional measures (SCIM, SCIM self-care subscore, and prehension). Functional measures were defined as the response variables and GRASSP subtests as the covariates. In addition, specific muscles within the MMT were also compared to functional measures including quantitative prehension tasks, again using general linear modeling. The strength of observed relationships between GRASSP impairment components and functional measures were used to exclude items and tests from the final GRASSP.

Preliminary components of the GRASSP were retained if there was a significant association with one of the three functional measures (SCIM, SCIM self-care subscore, and prehension). Strength of association was established by the p value. A p value $\leq .05$ was considered significant and $\leq .10$ approaching significance. General linear modeling results are summarized in **Table 3**. There were no significant associations between tone (Ashworth), SWM dorsal sensation, and S2PD with the three response (functional) variables. These elements were subsequently eliminated from the GRASSP. The most significant associations were found for strength, SWM palmar sensation, and grasp function. Pearson correlation coefficients were conducted between S2PD and quantitative prehension tasks. Individual neurological levels (C6 and C7) showed weak, although significant, correlations with the three fine motor tasks of quantitative prehension (Task 3, 0.496; Task 5, 0.388; Task 6, 0.355; $p < .001$). The results of the linear modeling and poor correlations with prehension tasks justified removal of S2PD from the GRASSP.

In addition to applying general linear

Table 3. Modeling for GRASSP components

Response variables	Step 1		Step 2		Step 3		Step 4	
	Covariates	***p***	**Covariates**	***p***	**Covariates**	***p***	**Covariates**	***p***
Prehension total	Strength	*.0001*	Strength	*.0001*	Strength	*.0001*	Strength	*.0001*
	Tone	.1516	Tone	.1376	SWM-D	.5229	SWM-P	*.0729*
	SWM-D	.6785	SWM-D	.6825	SWM-P	*.0613*	Tone	.1156
	SWM-P	.1516	SWM-P	.1456				
	S2PD	.9279						
SCIM self-care (subscore)	Strength	.1413	Strength	.1372	Strength	.1349	Strength	.1011
	Tone	.9115	Tone	.8783	SWM-D	.4464	SWM-P	*.0280*
	SWM-D	.5064	SWM-D	.4414	SWM-P	.3865	Qn-Grasp	*.0289*
	SWM-P	.5122	SWM-P	.4198	Qn-Grasp	*.0259*		
	S2PD	.8266	Qn-Grasp	.0316				
	Qn-Grasp	*.0330*						
SCIM total	Strength	.3028	Strength	.3090	Strength	.3793	Strength	*.0002*
	Tone	.3328	Tone	.3991	SWM-D	.3177	Qn-Grasp	*.0098*
	SWM-D	.1974	SWM-D	.2692	SWM-P	.6852		
	SWM-P	.8253	SWM-P	.5641	Qn-Grasp	*.0059*		
	S2PD	.4212	Qn-Grasp	*.0103*				
	Qn-Grasp	*.0103*						

Note: Additional combinations of linear regression were conducted to ensure that the combinations presented in this table were the most optimal. GRASSP = Graded Redefined Assessment of Strength, Sensibility and Prehension; SWM-D = Semmes Weinstein monofilaments-dorsal; SWM-P = Semmes Weinstein monofilaments-palmar; S2PD = static two-point discrimination; Qn = quantitative.

modeling to determine which subtests to retain in the final GRASSP, a similar method was used to determine which individual muscles from the MMT should be retained based on the strength of association to function. Functional measures were again used as the response variables and individual muscles from MMT as the co-variates. A rating system was devised where individual muscles scored 1 for every significant association ($p \leq .05$) to a response variable and a 0.5 if a relationship approached significance ($p \leq .10$). Nine associations were evaluated for each individual muscle, and scores were summed for a maximal possible score of 9 (**Table 4**). Individual muscles were eliminated if their rating was less than 1. Based on the regression analysis, 10 muscles had a rating 1 or above. One muscle, the abductor policis brevis, was eliminated. Wrist extension only approached significance for the SCIM self-care subscore; however, a decision was made to retain it in the final GRASSP due to its role as a key muscle in the ISNCSCI.

Summary and Future Steps

The GRASSP was conceived as an impairment measure for the upper limb that would be useful for assessing subtle neurological changes post cervical SCI during the acute, subacute, and postacute phases. Currently there is no validated and widely accepted measure for assessing the upper limb following cervical SCI. The preliminary work, presented in this article, successfully demonstrated a relationship among components of the GRASSP, measuring impairment, and function. To substantiate the efficacy and use of experimental agents for enhancing neurological recovery, future investigators will need to demonstrate both a change in impairment and a meaningful change in function through a responsiveness study. The GRASSP was developed to fill this gap and facilitate the performance of future clinical trials.

Based on the results of our analyses, the preliminary GRASSP was modified to maximize the link between impairment and function. Static two-point discrimination, magnitude of tone (Ashworth), and one muscle (abductor policis brevis) failed to demonstrate significant associations between impairment and function. These items were subsequently eliminated from the GRASSP. The current version (Version I) consists of SWM, MMT (10 muscles), and prehension testing. The development of the GRASSP represents one of the first steps to develop an upper limb impairment measure for SCI based on a large cohort of data.

The findings presented in this article are a small part of a larger, ongoing study designed to establish the reliability and validity of the GRASSP. A longitudinal study that will analyze the results of repeated measures of the GRASSP and functional measures of change on the same individuals over the course of a year will be undertaken. The results will provide data for responsiveness of the test, a recovery profile of the upper limb, and minimal clinically important differences of the upper limb for the tetraplegic population.

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Table 4. Relationship between individual muscles and functional tests/tasks of quantitative prehension

Co-variates / Muscles	Response variables: Prehension total	SCIM Self-care subscore	SCIM total	Task 1	Task 2	Task 3	Task 4	Task 5	Task 6	Rating /9
S1	.410	.3791	*.0080*	.6051	.3327	.9503	.4302	.2396	.7147	1
S2	.220	.7811	*.0014*	.2101	.7110	.1029	.1016	.3724	.4301	1
S3	*.0398*	.7698	.1722	.1337	*.0047*	*.0165*	.1155	.4137	.8227	3
S4	.8755	*.0658*	.1584	.9102	.4919	.3162	.4095	.6752	.5987	0.5
S5	.0795	*.0180*	*.0003*	.1751	.0947	.9013	*.0934*	*.0729*	*.0720*	3.5
S6	.349	.4162	*.0050*	.9536	.1276	.4458	.3365	.1987	.8850	1
S7	*.0015*	.5595	.8152	*.0878*	*.0110*	*.0209*	*.0059*	*.0007*	*.0974*	5.5
S8	.1886	*.0638*	*.0477*	.3646	.4000	.1797	.1631	.1089	.9554	1
S9	.8655	*.0881*	*.0458*	.4956	.8268	.8164	.4980	.4455	.9053	1.5
S10	.4318	.7081	.3687	.5067	.2313	.8020	.3457	.6691	.2656	0
S11	*.0188*	*.0656*	.4723	*.0233*	*.0320*	*.0694*	.2521	*.0520*	.1742	4.5

Note: Underlined values indicate muscles with significant or approaching significance association to function. SCIM = Spinal Cord Independence Measure. S1 = anterior deltoid; S2 = elbow extensor; S3 = elbow flexor; S4 = wrist extensor; S5 = extensor digitorum; S6 = flexor digitorum; S7 = flexor policis longus; S8 = abductor digiti minimi; S9 = first dorsal interossei; S10 = abductor policis brevis; S11= opponens policis.

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APPENDIX

Modifications to the Sollerman Hand Function Test

The zero to four scoring scale was increased to zero to five, terminology was modified, and timing was eliminated. All tasks requiring both hands were eliminated. Below summarizes all of the modifications made to the SHFT to design the performance based prehension testing for the GRASSP.

Sollerman Hand Function Test[4]	**Modified Sollerman Hand Function Test**
20 tasks	6 tasks
1 to 3 tasks for each grip	1 to 2 tasks per grip
7 grips: pulp pinch, lateral key pinch, tripod pinch, five finger pinch, spherical grasp, diagonal grasp, transverse grasp	5 grips: pulp pinch, lateral key pinch, tripod pinch, spherical grasp, transverse grasp
5-point scale	6-point scale
Incorporated bilateral tasks	Unilateral tasks only
Functional tasks	Functional tasks
Timed each task	Eliminated timing as part of scoring

Scoring (a maximum of 1 minute and 15 seconds is allowed for each task)

0 - the task cannot be conducted at all

1 - the task cannot be completed (less than 50% of the task) and the expected grasp is not used

2 - the task is not completed (50% or more of the task) and the expected grasp is not used

3 - the task is completed using tenodesis or an alternative grasp other than the expected grasp

4 - the task is completed using the expected grasp with difficulty (lack of smooth movement or difficult slow movement)

5 - the task is completed without difficulties using expected grasping pattern and unaffected hand function

Note: 50% of Task 1 is when the participant has begun to pour the water, 50% of Task 4 is when the participant is able to get the key to insertion point.

Validation of the electrical perceptual threshold test as a quantitative assessment of cutaneous sensory function for spinal cord injury trials

PI: Professor Peter Ellaway, Dept of Clinical Neuroscience, Imperial College, London, UK

Background: The current gold standard for clinical assessment of spinal cord injury (SCI) is the American Spinal Injuries Association (ASIA) Impairment Scale for sensory function. There are limitations to the ASIA assessment: (1) it uses an ordinal rather than a quantitative scale, (2) there is a strong component of subjectivity, (3) evaluation of each dermatome is scored simply as either normal, absent or abnormal (including both heightened and lowered sensitivity). Improved outcome measures should allow both for improvements and worsening of the condition of those undergoing clinical trials for SCI, and be capable of detecting change at a single vertebral level of the spinal cord. The Electrical Perceptual Threshold test (EPT) (Belci et al, 2004) meets these criteria.

Electrical Perceptual Threshold: EPT provides a quantitative and more objective measure of threshold for cutaneous sensory function for each dermatome. The method uses incrementing electrical stimulation and the method of limits to determine threshold. It has been validated against the AIS sensory grading in SCI (Ellaway et al, 2004; Savic et al, 2006) and undergone repeatability evaluation for inter and intra-rater trials in SCI (King et al, 2009). Good reliability has now been confirmed (Leong et al, 2009) in control subjects and validation provided against dermatomal somatosensory evoked potentials in SCI (Kramer at al, 2009). It is well tolerated by subjects, not time consuming to perform and requires apparatus of relatively modest cost. Little training of operators is required to obtain reliable and repeatable measures.

Proposed project: To validate EPT against monofilaments (Semmes-Weinstein) in neurologically normal subjects. The first task force meeting of the NACTN group (Louisville, May 2009) identified the need to further validate the EPT test by comparing sensitivity to that of cutaneous mechanical stimulation using monofilaments. The proposed study would measure EPT and monofilament threshold for a set of dermatomes (C4, T1, T8 & L4) known to have a range of sensitivities in neurologically normal subjects. Twenty men and twenty age-matched women would be recruited for the study. They would be studied on two occasion s with an interval of at least one week, to provide additional intra-rater repeatability measures. Correlations between EPT and monofilament readings would be established. The study would be carried out in the UK laboratory led by Professor Ellaway employing a research assistant (part-time). The laboratory is already equipped with the apparatus required for the study, including a computer controlled stimulator (Digitimer DS5), Laptop PC and sets of Semmes-Weinstein monofilaments. A DAQ card interface is required to replace an obsolete model currently on loan to the laboratory.

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Costing for ***UK-based project:*** *To validate EPT against monofilaments (Semmes-Weinstein) in neurologically normal subjects.*

Support requested based on exchange rate £1 = $1.60

Item	Duration	Cost in £ sterling	Estimate in US$
Research assistant (0.4FTE*) Based on £35k p.a. + 26% NI**	6 months	8,800	14,080
Software development (@ £37.50 per hour)	16 hours	600	960
Travel costs (2x £15 x 40 subjects)		1,200	1,920
Travel costs staff (24 weeks @ £22)		528	845
Consumables: Electrodes, alcohol swabs		400	640
Equipment: Medical waveform generator interface (NI USB 9263)		428	685
Office and PC costs		200	320
TOTAL		12,156	**19,450**

*FTE, full-time equivalent. **NI, National Insurance

Justification for support

- 0.4 of a full-time equivalent research assistant is required to recruit subjects, perform EPT and collate results. Mrs Maria Catley (research technician) would be available to assume this post in February 2010. She has worked as Professor Ellaway's technician for many years, has the necessary skills and would require no training. The cost is based on her current salary.
- Equipment: Professor Ellaway will supply the stimulator (Digitimer DS5 - value $10,500) and Laptop PC required for the study. A Medical Waveform Generator interface (National Instruments NI USB 9263) is required to replace the current obsolete system on loan to the laboratory. Support is requested to expand the software need to accommodate the change in interface connection.
- Consumables: these are estimates based on past research and adjusted to the numbers of subjects and sessions envisaged for the project.
- Travel costs: These are based on the average cost of public transport (estimated from previous research) required to attend the laboratory.

Peter Ellaway 07 Jan 2010

Natural progression and recovery of cardiovascular parameters following traumatic spinal cord injury.

PI: Dr. Andrei Krassioukov, Vancouver, BC
Co-Investigators:
Dr. S Harkema, Louisville KY,
Dr. G. Guest
Dr. Grossman

Rationale:

Neurogenic shock is one manifestation of the dysfunctions of the autonomic nervous system observed following spinal cord injury (SCI) and manifests by a significant decrease in arterial blood pressure from baseline, and in cervical injuries, can result in severe hypotension and bradycardia [1-3]. There is some evidence suggesting that this event is more profound and long lasting in humans after SCI than in experimental animals. Neurogenic shock is most probably an effect of the imbalance in autonomic control, with an intact parasympathetic influence via the vagal nerve and a loss of sympathetic tone due disruption of supraspinal control. There are only few observations with small number of individuals suggesting that prolonged and severe hypotension, requiring vasopressive therapy, is associated with the severity of the SCI, and can last up to 5 weeks after injury [1;4]. In one study, Glenn et al. reported that severe hypotension was present in all 31 tetraplegic subjects assessed with severe SCI, half of whom required pressor therapy in order to maintain adequate arterial blood pressure [5]. In addition to the pronounced hypotension described, many patients with acute SCI experience severe abnormalities in heart rate. Bradycardia was reported in 64% to 77% of patients with cervical SCI during the acute post-injury stage, and was more severe and frequent within the first 5 weeks after injury [6-8]. Interestingly, Furlan and colleagues reported that the hypotension and bradycardia observed initially after injury persisted in the individuals with more severe injury of the descending cardiovascular autonomic pathways[9]. Moreover, all individuals in this group required vasopressor therapy in order to maintain systolic arterial blood pressure above 90 mmHg. In contrast, individuals with less severe injury to the descending cardiovascular pathways tended to show higher levels of blood pressure and heart rate, although minor and short-term hypotension and low heart rates were occasionally observed.

In addition to neurogenic shock, the acute phase of SCI is also associated with "spinal shock"[10-12]. Some authors use these terms interchangeably, however, it is important to recognize that these are two clinically important and distinct conditions. Neurogenic shock is characterized by changes occurring in blood pressure and heart rate (autonomic) control following SCI [1;3;4] whereas spinal shock is characterized by a marked reduction or abolition of motor and reflex function below the level of injury [12]. Clinically, spinal shock in humans can persist for days to weeks, with a mean duration of between four to six weeks after the injury. Traditional views of the clinical course of the recovery of spinal shock were related to the emergence of certain groups of reflexes [12]. For example, some considered that spinal shock had ended when the appearance of initial reflexes such as the bulbocavernosus reflex occurred in the first few days after

SCI, others with the recovery of deep tendon reflexes at two weeks post injury, while some groups classified the end of spinal shock as when the bladder reflex recovered after approximately two months. For further details, we refer readers to the recent work of Ditunno and colleagues [12].

Low arterial blood pressure (BP) and the presence of the neurogenic shock (BP below 90 mmHg) after SCI result in ischemia of the spinal cord and are major contributing factors to the cascade of the secondary mechanisms involved in further damage of fragile neuronal tissue [13;14]. Hypoperfusion of the spinal cord could result from both low systolic BP (SBP) and mechanical compression of the spinal cord [15-17]. Increases in systemic blood pressure may improve perfusion to the injured, distorted spinal cord [18-20]. Several contemporary series of spinal cord injured patients treated with aggressive medical management with maintenance of mean arterial blood pressure to high normal ranges (85 mmHg to 90 mmHg) have suggested improved neurological outcomes with this management plan [20]. Presently, there is no consensus for how long is vassopressive therapy should continue, at what level of arterial blood pressure it should be discontinued and whether it is better to achieve surgical decompression of the spinal cord early or to postpone the procedure until patients are more stable [21-23].

Presently we have well established and detailed criteria for the blood pressure parameters in able-bodied population and guideline for the monitoring of changes in these parameters[24]. For example criteria for hypertension in able-bodied individuals differ between clinic blood pressure, 24-h ABP and home blood pressure. Latest Guidelines for clinical evaluations of blood pressure suggest that a clinic blood pressure of >140/90mmHg, a home blood pressure of >135/85mmHg and a mean 24-h ABP of >130/80mmHg are regarded as indicators of hypertension. A normal home blood pressure for the able-bodied individual is <125/80mmHg. Unfortunately, at the present time we do not have a full clinical picture of the changes in cardiovascular parameters in individuals with SCI. Furthermore, we still do not appreciate the full extent of influence of the changes in arterial blood pressure on extend of neurological recovery following this devastating injury.

Aims:

Aim 1. To establish a data base with the natural progression and recovery of cardiovascular parameters in individuals with SCI.

Aim 2. To establish the effect of the changes in arterial blood pressure on potential neurological recovery following traumatic SCI.

Aim 3. To develop a guidelines on the acute monitoring and management of cardiovascular parameters for individuals with SCI.

Methods:

1. Each participating center will adhere to the established protocol for evaluation and recordings of cardiovascular parameters for individuals with SCI.
2. The use of standardized equipment and trained personnel will be crucial for the outcomes of the study.

Protocols and Data Collection:

Demographics:

1. Age
2. Sex
3. Education
4. Ethnicity

SCI related data:

1. Time and date of injury
2. Causes of injury
3. Initial assessment at the site of injury: LOC, additional injuries, initial neurological examination.

SCI surgical data:

1. Date and extend of the spine/spinal cord surgical procedure
2. Dates and extend of secondary surgical non spine procedures.

Cardiovascular parameters (Timing and documentation of methods of measurements): Parameters will include systolic and diastolic arterial blood pressure and heart rate.

*Cardiovascular parameters **prior the admission to the tertiary** medical centre:*

1. At the site of injury – records from paramedics reports
2. At the time of admission to the first ER/Hospital
3. Time of initiation of the fluid resuscitation or/and vasopressor therapy

*Cardiovascular parameters **at the tertiary** (final treatment) centre:*

4. At the time of admission to the tertiary hospital ER.
5. Time of initiation of the fluid resuscitation or/and vasopressor therapy
6. Daily morning (7:00-8:00 am) cardiovascular parameters from day 0-30 days post injury. (*We still have to make decision what to do with records if continuous blood pressure recordings are available. This will require more intense timing for the charting or transferring the data into the data base*! This will be made during the training team meeting!!!!!)
7. Timing of discontinuation of the vasopressors support!
8. Date of initial mobilization
9. Orthostatic hypotension presence, severity and duration.

Documentation of ASIA Impairment Scale:

1. Admission to the tertiary centre – Day 0
2. Day 3, Day 7, Day 30, Day 90, Day 180, Day 365.

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Specific Aims: The primary goal of this research is to cultivate an objective neurophysiological measurement tool that assesses motor and sensory neural recovery in individuals with SCI for use in multi-center clinical trials.

The proposed project will evaluate the BMCA and EPT by comparing parameters calculated from surface electromyographic (sEMG) activity to scores on the American Spinal Injury Association (ASIA) Impairment Scale (AIS), a subjective clinical assessment which utilizes manual muscle testing and subjective sensation of normal, absent or abnormal sensation .

We are requesting funds in order to develop clinically applicable hardware and software for testing reliability and sensitivity of the BMCA and EPT in persons with SCI.

Specific Aim 1: To develop standardized instrumentation and software with standard interface for analyses and reporting.

Specific Aim 1a: To develop BMCA standard protocol for total body [upper, lower, and trunk]

Specific Aim 1b: To establish protocols for examiners for clinical sites on standardized hardware and software set up and BMCA and EPT testing protocols

Specific Aim 2: To acquire data from 10 non-injured and 10 SCI patients using the standardized hardware and software and clinical protocols at the UofL NACTN site.

Early versus Late Surgical Decompression for Traumatic Spinal Cord Injury: Results of the Surgical Trial in Acute Spinal Cord Injury Study (STASCIS)

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Key words: Spinal Cord Injury, Surgical Timing, Neurologic Outcomes, Complications

Abstract

Background:

There is convincing experimental evidence in animal models that early decompressive surgery in the setting of spinal cord injury (SCI) attenuates secondary injury mechanisms and improves neurologic outcomes. However, the effect of early surgical decompression in patients with an acute SCI remains uncertain.

Methods:

We have performed a multicenter, international, prospective controlled study (Surgical Trial in Acute Spinal Cord Injury Study: STASCIS) in adults aged 16-70 with a cervical SCI , to evaluate the impact of early (<24 hours after injury) or late (≥24 hours after injury) decompressive surgery. The primary outcome of interest was AIS grade change at 6 month follow-up. Secondary outcomes included an assessment of rates of mortality and secondary complications.

Results:

A total of 313 patients with acute cervical spinal cord injury were enrolled in the study. Of the 313 study participants, 182 underwent "early" surgery, at a mean of 14.21(±5.44) hours, with the remaining 131 having underwent "late" surgery, at a mean of 48.37(±29.25) hours ($p<0.0001$). At 6 months follow-up, 19.9% of patients undergoing early surgery showed a ≥2 grade change in AIS as compared to 8.8% in the late decompression group ($p=0.036$). The multivariate analysis, adjusted for baseline AIS and age, demonstrated greater AIS grade improvements in those treated with early as compared to late surgery($p=0.04$). During the 30 day post injury period, there was 1 mortality in both the early and late surgery groups. Major inpatient complications occurred in 24.2% of early surgery patients and 30.5% of late surgery patients($p=0.21$).

Conclusion:

Early decompressive surgery after cervical SCI can be performed safely and results in improved neurologic outcome as measured by AIS grade conversion. Moreover, early surgery may result in reduced rates of major complications.

Riluzole Clinical Phase I Trial Human Plasma Data Input

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Table I Patient Enrolled and Samples Received Time Form Time for Riluzole Phase I Clinical Trial

Subject ID	Enrolled Time	Enrolled Sites	Received Time	Detailed Description	Memo
R07-0001	April 13, 2010	Maryland	April 28, 2010	Riluzole was given 11.5 hours after post-injury. Medication ingested N.G. 00:30 AM on April 13, 2010.	Day 3: dosing time 10:25 and 22:00 on April 15, 2010; Day 14: dosing time 09:45 on April 26, 2010
R07-0002	May 1, 2010	Maryland	May 18, 2010	Riluzole was given 9 hours post-injury. Medication ingested P.O. 11:30 AM on May 1, 2010.	Day 3: dosing time 09:32 and 22:00 on May 3, 2010; Day 14: dosing time 09:00 and 21:00 on May 14, 2010
R07-0003	May 8, 2010	Maryland	May 27, 2010	Riluzole was given 8 hours post-injury. Medication ingested P.O. 14:36 PM on May 8, 2010.	Day 3: dosing time 13:26 and 20:55 on May 10, 2010; Day 14: dosing time 10:05 and 22:00 on May 21, 2010
R07-0004	May 31, 2010	Maryland	June 17, 2010	Riluzole was given 4.5 hours post-injury. Medication ingested P.O. 15:15 PM on May 31, 2010.	Day 3: dosing time 21:05 pm on June 2nd, 2010; Day 14: dosing time 10:00 am on June 13, 2010.
R07-0005	June 20, 2010	Maryland	July 9, 2010	Riluzole was given at 11:10 (first dose), baseline was withdrawn at 11:00.	Day 3: dosing time 21:45 pm on June 22, 2010; Day 14: dosing time 10:00 am on July 3, 2010.
R07-0006	July 3, 2010	Maryland	July 30, 2010	Riluzole was given at 18:30, 7 hours from injury on July 3, 2010.	Day 3: dosing time 22:00 pm on July 5, 2010; Day 14: dosing time 10:10 am on July 16, 2010.
R07-0007	July 9, 2010	Maryland	July 30, 2010	The first dose time for R07-0007 is 0630.	Day 3: dosing time 15:20 pm on July 11, 2010; Day 14: dosing time 10:15 am on July 22, 2010.
R07-0008	September 12, 2010	Maryland	September 30, 2010	The first dose time for R07-0008 is 0430.	Day 3: dosing time 10:30 am on September 14, 2010; Day 14: dosing time 10:02 am on September 25, 2010.

R10-0001	July 4, 2010	Thomas Jefferson	July 20, 2010	He is a 22y/o with a C6 burst fracture, ASIA C. He started Riluzole on 7/4/2010 at 23:30. His injury was 7/4/2010 at 13:00.	Day 3: dosing time 09:00 am on July 7, 2010; Day 14: dosing time 09:00 am on July 17, 2010. On both Day 3 and 14, dosing times are 09:00 and 21:00.
R10-0002	July 15, 2010	Thomas Jefferson	July 30, 2010		Day 3: dosing time 09:00 am on July 17, 2010; Day 14: dosing time 09:00 am on July 28, 2010. On both Day 3 and 14, dosing times are 09:00 and 21:00.
R10-0003	September 5, 2010	Thomas Jefferson	September 25, 2010	He is an 18 y/o male that was ejected from a truck after hitting a pole; T12 ASIA A. First Dose for patient R10-003 is 1150 on 09/05/2010.	Day 3: dosing time 09:00 am on September 7, 2010; Day 14: dosing time 09:00 am on September 18, 2010. On both Day 3 and 14, dosing times are 09:00 and 21:00.
R10-0004	September 8, 2010	Thomas Jefferson	September 25, 2010	He is 20 y/o male involved in a motorcycle accident; T12 ASIA A; His accident was at 2100 last night and first Dose for patient R10-004 is 0500 on 09/08/2010.	Day 3: dosing time 09:00 am on September 10, 2010; Day 14: dosing time 09:00 am on September 21, 2010. On both Day 3 and 14, dosing times are 09:00 and 21:00.
R10-0005	September 12, 2010	Thomas Jefferson	September 30, 2010	He is 65y/o male; dense central cord C5 ASIA B; first dose of study drug was given at 0035 on 09/12/2010.	Day 3: dosing time 09:00 am on September 14, 2010; Day 14: dosing time 09:00 am on September 25, 2010. On both Day 3 and 14, dosing times are 09:00 and 21:00.
R10-0006	September 13, 2010	Thomas Jefferson	September 30, 2010	He fell down the stairs; C4 ASIA A; first dose was given at 19:50 on 09/13/2010.	Day 3: dosing time 09:00 am on September 16, 2010; Day 14: dosing time 09:00 am on September 27, 2010. On both Day 3 and 14, dosing times

					are 09:00 and 21:00.
R10-0007	October 4, 2010	Thomas Jefferson		He started drug at 1850 last evening. He is a 41 year old male who fell from 15 feet and has a complete T9 ASIA A injury.	
R05-001	August 30, 2010	Virginia	September 25, 2010	He is 19 years old that was involved in MVA; T11-T12 ASIA A; Control blood sample obtained at 1:00 AM; First dose of Riluzole at 1:50 AM	Day 3: dosing time 09:00AM on September 2, 2010
R05-002	September 2, 2010	Virginia	September 25, 2010	She is 37 years old female that was involved in MVA; T3-T4 ASIA A; Control blood sample obtained at 11:55 AM (Pregnancy test negative); First dose of Riluzole at 12:44 PM	Day 3: dosing time 12:48 pm on September 5, 2010; Day 14: dosing time 12:30 am on September 15, 2010.
R05-003	September 2, 2010	Virginia	September 25, 2010	He is 25 years old male that was involved in MVA; C4-C5 ASIA B; Control blood sample obtained at 09:55 AM; First dose of Riluzole at 10:00 AM	Day 3: dosing time 15:50 pm on September 6, 2010; Day 14: dosing time 14:35 am on September 17, 2010.
R05-004	September 22, 2010	Virginia		He is 53 y male who fell 70 to 100 feet while working on a bridge ;C2 to C5 - ASIA A; First Riluzole dose was given via NG tube at 16:55	
	October 4, 2010	UT		The patient is an 18 y/o male, C5-7 incomplete, injured Monday evening at 11:43 PM. The study drug was given 11 hours post injury.	

Table II Riluzole Phase I Clinical Trial Human Pharmacokinetic Data Input

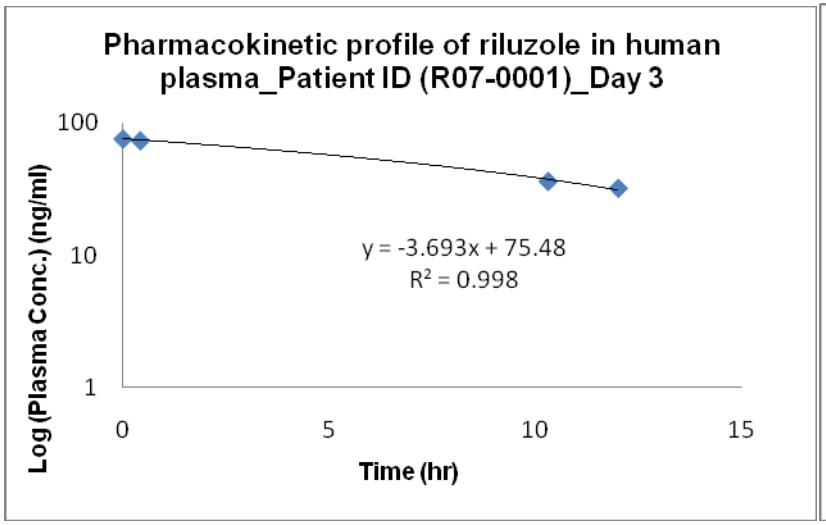
Subject ID		Dosing Number	C_{max} (ng/ml)	C_{peak} (ng/ml)		C_{min} (ng/ml)	C_{trough} (ng/ml)	
				Individual Conc.	Mean ± SD (CV%)		Individual Conc.	Mean ± SD (CV%)
R07-0001	Day 3	6# dose		74.26	73.66 ± 1.29 (2.0)		36.03	36.03 ± 0.77 (2.0)
			75.91	72.18		32.15	35.26	
				74.54			36.80	
	Day 14	28# dose		27.69	28.52 ± 0.77 (3.0)			
			29.20	28.43		5.50	7.57	
				29.43				
R07-0002	Day 3	5# dose		66.68	68.25 ± 1.82 (2.7)		36.56	37.03 ± 0.43 (1.2)
			70.37	70.25		33.80	37.15	
				67.83			37.40	
	Day 14	27# dose		67.26	68.12 ± 0.91 (1.3)		18.67	16.79 ± 1.73 (10.3)
			70.88	68.04		12.91	16.45	
				69.07			15.26	
R07-0003	Day 3	6# dose		115.75	115.81 ± 0.14 (0.1)		20.87	20.28 ± 1.21 (6.0)
			126.55	115.98		15.09	18.89	
				115.72			21.08	
	Day 14	27# dose		38.62	35.68 ± 2.60 (7.3)		17.06	15.97 ± 0.95 (6.0)
			36.67	33.70		13.59	15.47	
				34.71			15.37	
R07-0004	Day 3	5# dose		150.36	147.73 ± 2.72 (1.8)		38.22	39.82 ± 1.77 (4.4)
			198.20	144.93		33.96	41.72	
				147.73			39.52	
	Day 14	26# dose		24.41	24.89 ± 0.44 (1.8)		12.67	12.75 ± 0.10 (0.8)
			30.70	25.26		12.60	12.86	
				25.00			12.72	
	Day 3	6# dose		96.68	97.31 ± 0.96 (1.0)		42.19	41.99 ± 0.45 (1.1)
			114.35	96.83		41.73	42.30	

R07-0005				98.41			41.48	
	Day 14	27# dose		30.35	31.92 ± 1.69 (5.3)		14.78	14.83 ± 0.26 (1.6)
			36.98	33.70		14.73	14.61	
				31.69			15.12	
R07-0006	Day 3	5# dose		140.64	144.07 ± 3.86 (2.7)		18.49	18.09 ± 0.34 (1.9)
			233.43	148.24		16.19	17.91	
				143.32			17.88	
	Day 14	26# dose		26.88	26.85 ± 0.21 (0.8)		11.85	12.09 ± 0.41 (3.4)
			32.09	26.62		11.97	11.85	
				27.04			12.57	
R07-0007	Day 3	6# dose		45.81	47.20 ± 1.53 (3.2)		46.06	44.56 ± 2.12 (4.8)
			48.76	48.84		41.22	45.49	
				46.93			42.13	
	Day 14	27# dose					60.87	62.82 ± 1.93 (3.1)
			No samples	No samples			64.74	
							62.84	
R07-0008	Day 3	5# dose		94.74	99.44 ± 1.99 (2.0)		47.64	44.48 ± 0.33 (0.7)
			117.93	100.39		42.32	42.66	
				103.20			43.13	
	Day 14	27# dose		181.82	179.83 ± 4.20 (2.3)		12.48	12.73 ± 0.69 (5.4)
			295.13	175.86		12.50	13.34	
				181.81			12.37	
R10-0001	Day 3	6# dose		84.30	86.49 ± 3.97 (4.6)		40.64	40.05 ± 0.51 (1.3)
			102.06	91.08		39.27	39.70	
				84.09			39.81	
	Day 14	28# dose		32.82	34.10 ± 1.25 (3.7)		11.80	11.57 ± 1.03 (8.9)
			43.56	35.32		11.25	12.47	
				34.17			10.45	
R10-0002	Day 3	5# dose		50.03	49.27 ± 0.83 (1.7)		58.30	57.56 ± 1.08 (1.9)
			unavailable	49.41		unavailable	56.31	
				48.38			58.06	
	Day 14	27# dose		48.37	48.02 ± 0.69 (1.4)		26.10	25.07 ± 1.14 (4.6)
			55.63	48.47		24.66	23.83	

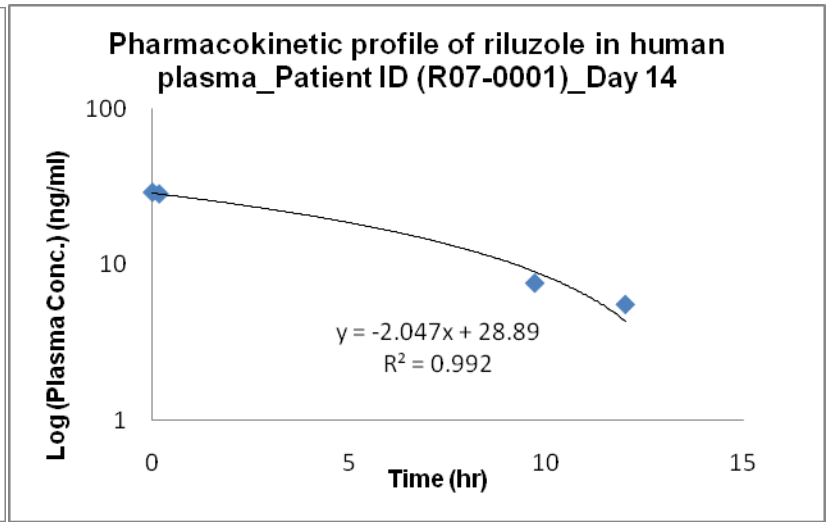
				47.23			25.28	
R10-0003	Day 3	5# dose		66.35	67.48 ± 0.17 (0.2)		53.01	54.75 ± 0.64 (1.2)
			71.51	68.16		54.14	56.07	
				67.92			55.16	
	Day 14	27# dose		42.00	40.47 ± 1.96 (4.9)		25.10	24.31 ± 0.93 (3.8)
			45.06	38.32		23.68	24.57	
				41.09			23.26	
R10-0004	Day 3	5# dose		141.58	142.73 ± 1.12 (0.8)		55.84	55.37 ± 2.58 (4.7)
			174.23	142.80		52.66	52.58	
				143.82			57.68	
	Day 14	27# dose		48.79	50.84 ± 2.20 (4.3)		25.35	24.61 ± 0.67 (2.7)
			59.23	53.16		23.68	24.48	
				50.58			24.02	
R10-0005	Day 3	6# dose						
	Day 14	28# dose						
R10-0006	Day 3	6# dose		176.98	174.00 ± 2.93 (1.7)		52.54	53.17 ± 1.91 (3.6)
			223.32	173.90		49.95	55.32	
				171.11			51.66	
	Day 14	28# dose		59.96	59.44 ± 0.54 (0.9)		13.97	12.70 ± 1.10 (8.7)
			82.26	58.88		11.70	12.11	
				59.49			12.03	
R05-001	Day 3	5# dose		130.08	128.47 ± 1.66 (1.3)		29.50	30.02 ± 0.63 (2.1)
			177.47	126.77		25.55	30.72	
				128.55			29.83	
. R05-002	Day 3	7# dose		103.21	106.53 ± 2.89 (2.7)		60.98	60.17 ± 1.13 (1.9)
			124.62	108.46		56.31	58.88	
				107.93			60.67	
	Day 14	29# dose		133.08	133.60 ± 1.07 (0.8)		62.32	63.76 ± 2.59 (4.1)
			157.46	134.83		58.72	66.75	

				132.90			62.22	
R05-003	Day 3	5# dose		183.46	186.95 ± 3.99 (2.1)		156.49	161.81 ± 4.68 (2.9)
			190.19	186.1		159.43	165.27	
				191.30			163.67	
	Day 14	27# dose		77.15	81.86 ± 4.24 (5.2)		20.84	20.86 ± 0.24 (1.1)
			110.92	85.37		17.92	21.11	
				83.01			20.64	

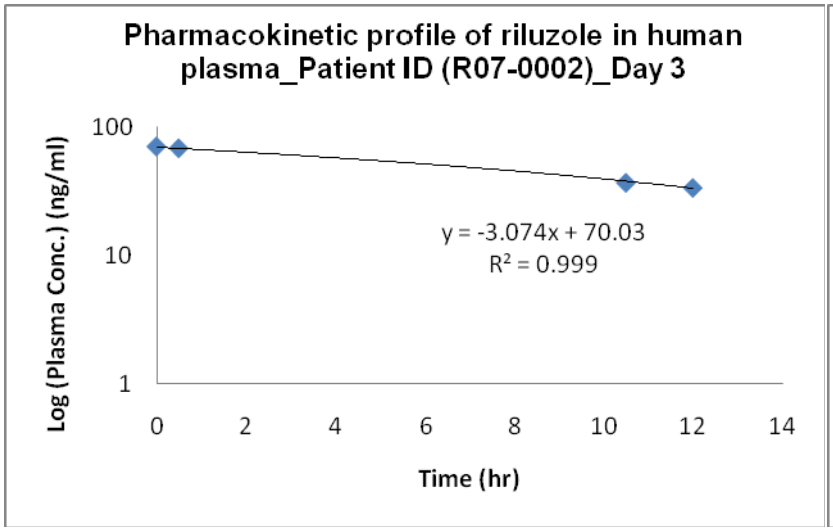
(A)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0001)_Day 3
Log (Plasma Conc.) (ng/ml)
100
10
1
0
5
10
15
Time (hr)
y = -3.693x + 75.48
R² = 0.998

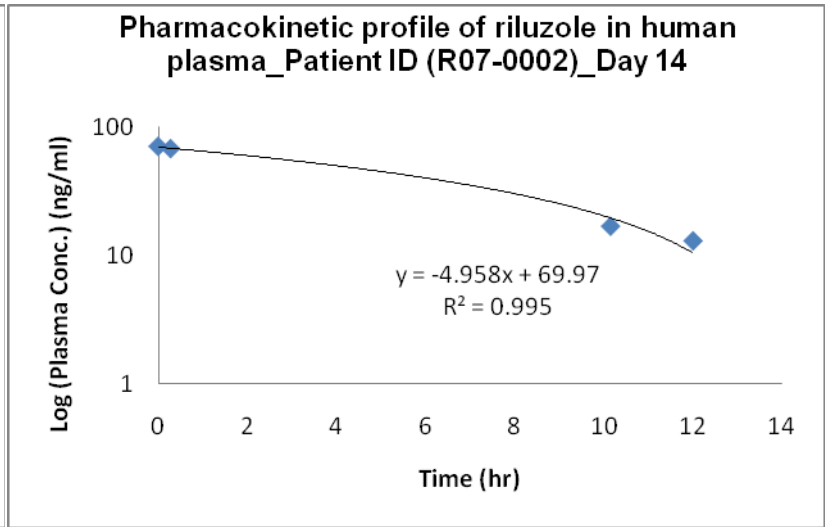
(B)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0001)_Day 14
Log (Plasma Conc.) (ng/ml)
100
10
1
0
5
10
15
Time (hr)
y = -2.047x + 28.89
R² = 0.992

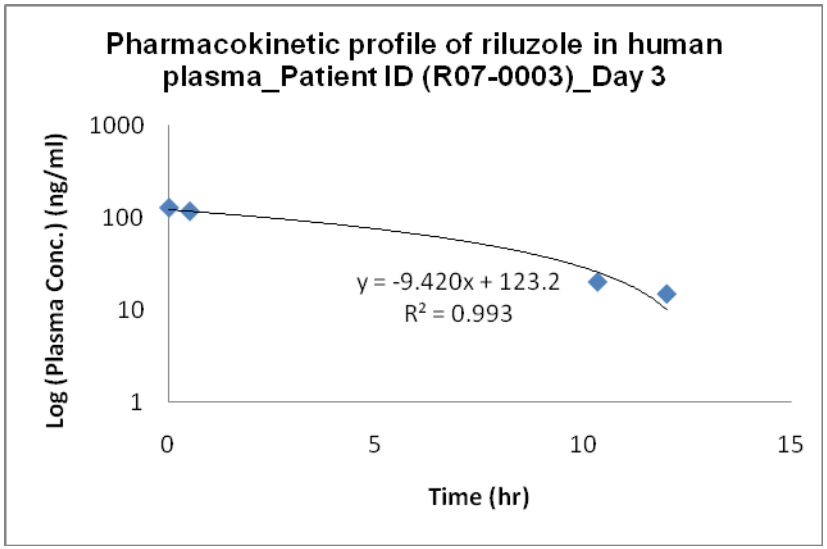
(C)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0002)_Day 3
Log (Plasma Conc.) (ng/ml)
100
10
1
0
2
4
6
8
10
12
14
Time (hr)
y = -3.074x + 70.03
R² = 0.999

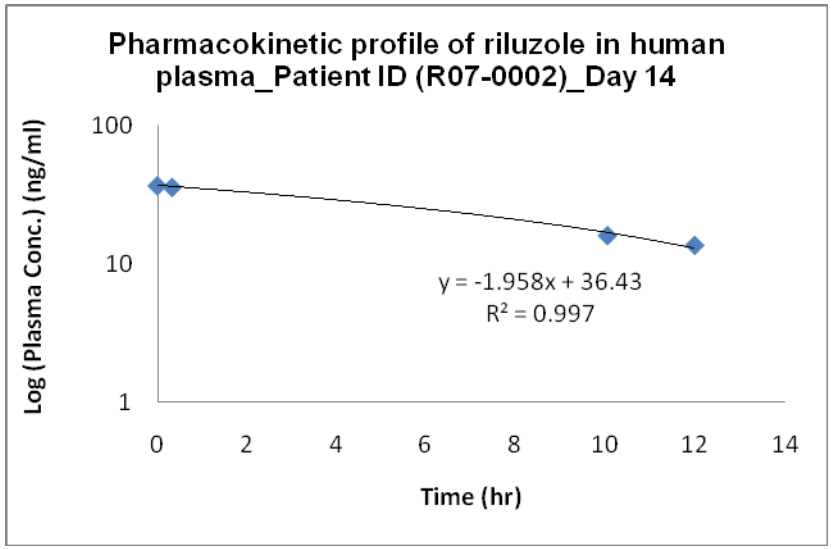
(D)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0002)_Day 14
Log (Plasma Conc.) (ng/ml)
100
10
1
0
2
4
6
8
10
12
14
Time (hr)
y = -4.958x + 69.97
R² = 0.995

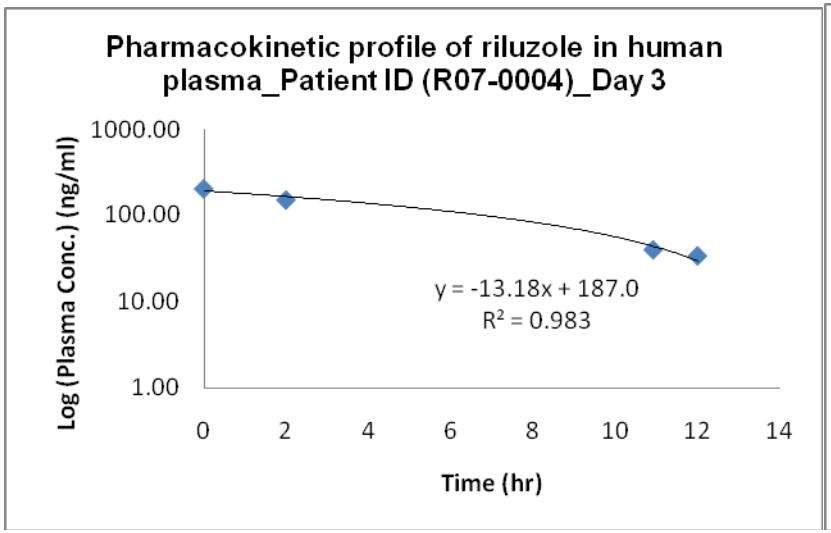
(E)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0003)_Day 3
Log (Plasma Conc.) (ng/ml)
1000
100
10
1
y = -9.420x + 123.2
R² = 0.993
0
5
10
15
Time (hr)

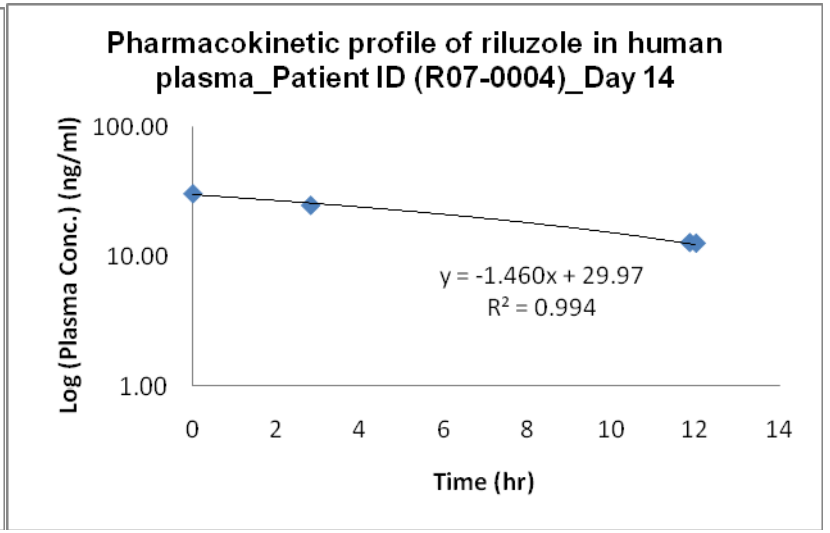
(F)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0002)_Day 14
Log (Plasma Conc.) (ng/ml)
100
10
1
y = -1.958x + 36.43
R² = 0.997
0
2
4
6
8
10
12
14
Time (hr)

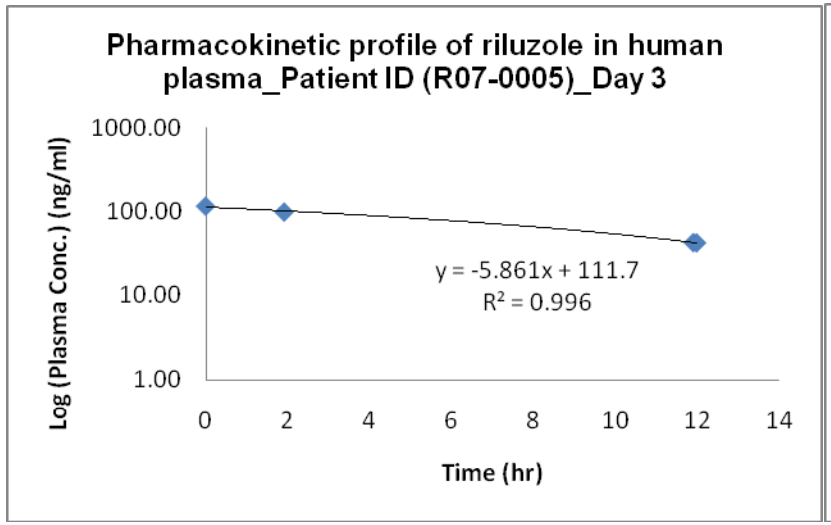
(G)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0004)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
y = -13.18x + 187.0
R² = 0.983
0
2
4
6
8
10
12
14
Time (hr)

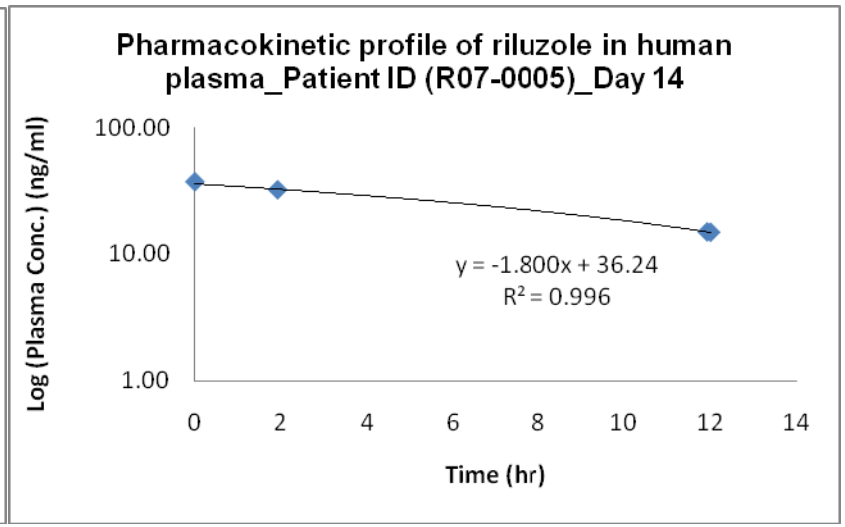
(H)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0004)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -1.460x + 29.97
R² = 0.994
0
2
4
6
8
10
12
14
Time (hr)

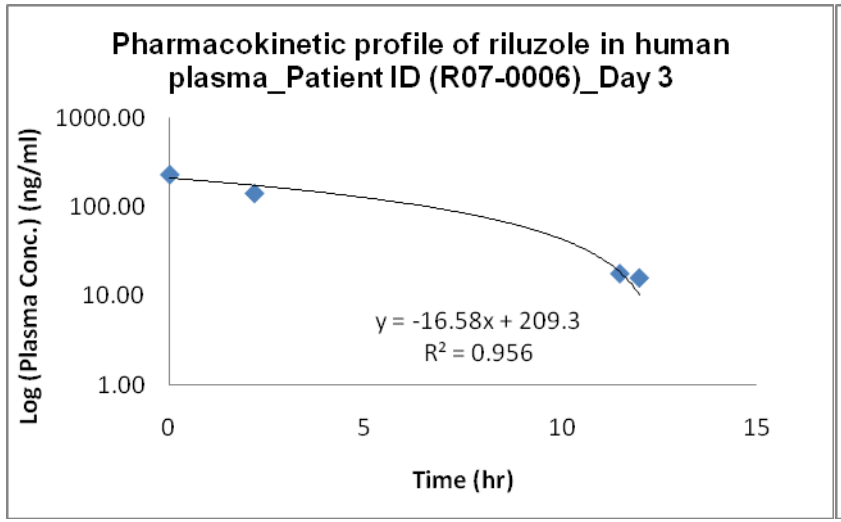
(I)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0005)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
y = -5.861x + 111.7
R² = 0.996
0
2
4
6
8
10
12
14
Time (hr)

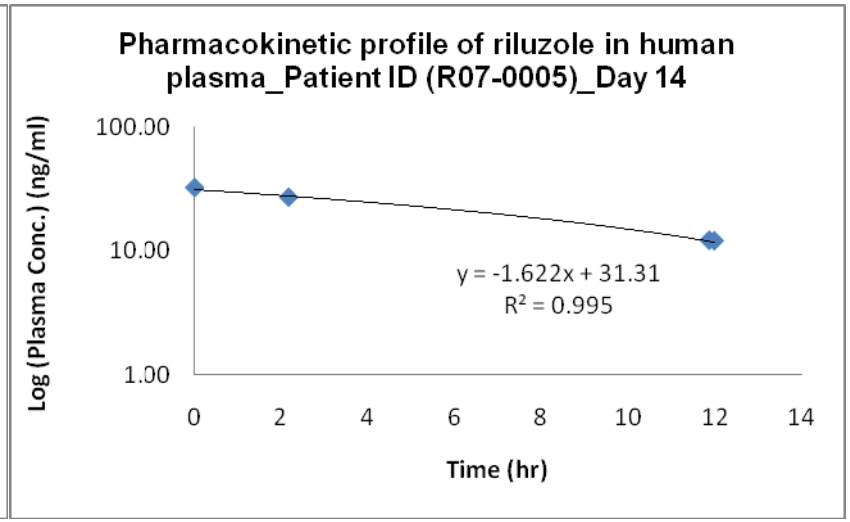
(J)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0005)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -1.800x + 36.24
R² = 0.996
0
2
4
6
8
10
12
14
Time (hr)

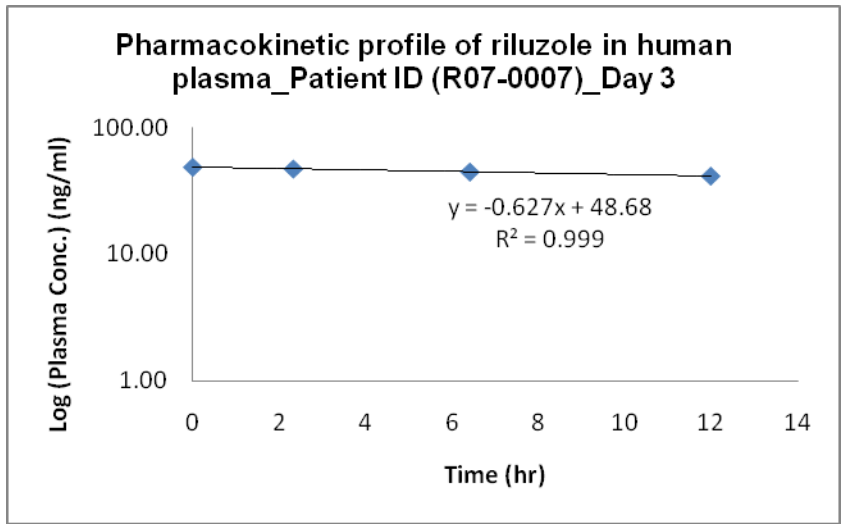
(K)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0006)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
y = -16.58x + 209.3
R² = 0.956
0
5
10
15
Time (hr)

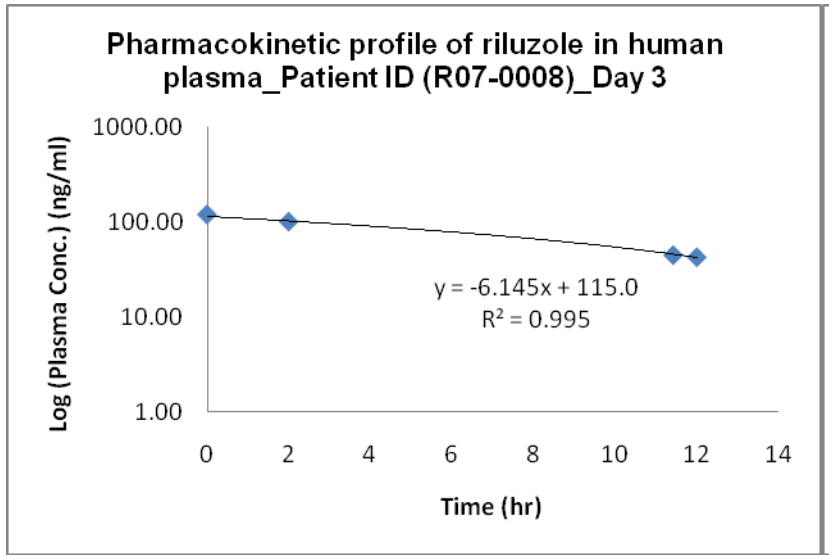
(L)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0005)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -1.622x + 31.31
R² = 0.995
0
2
4
6
8
10
12
14
Time (hr)

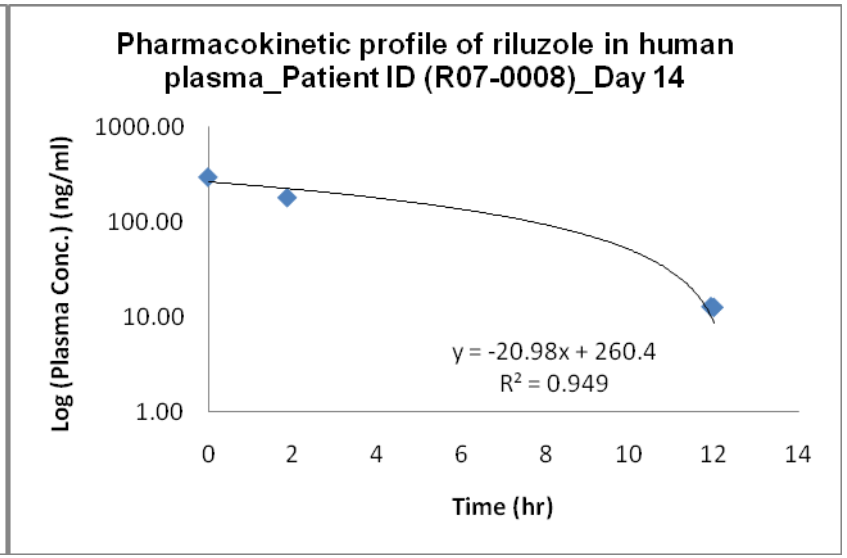
(M)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0007)_Day 3
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -0.627x + 48.68
R² = 0.999
0
2
4
6
8
10
12
14
Time (hr)

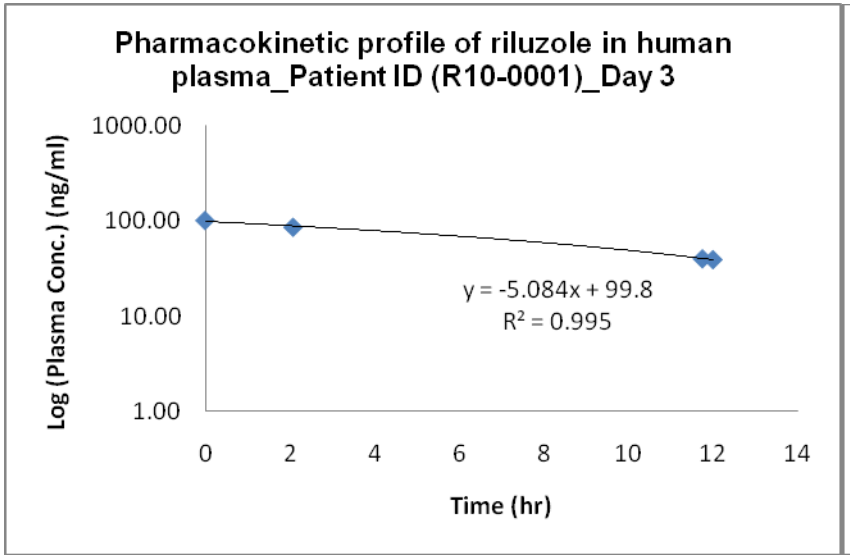
(N)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0008)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
y = -6.145x + 115.0
R² = 0.995
0
2
4
6
8
10
12
14
Time (hr)

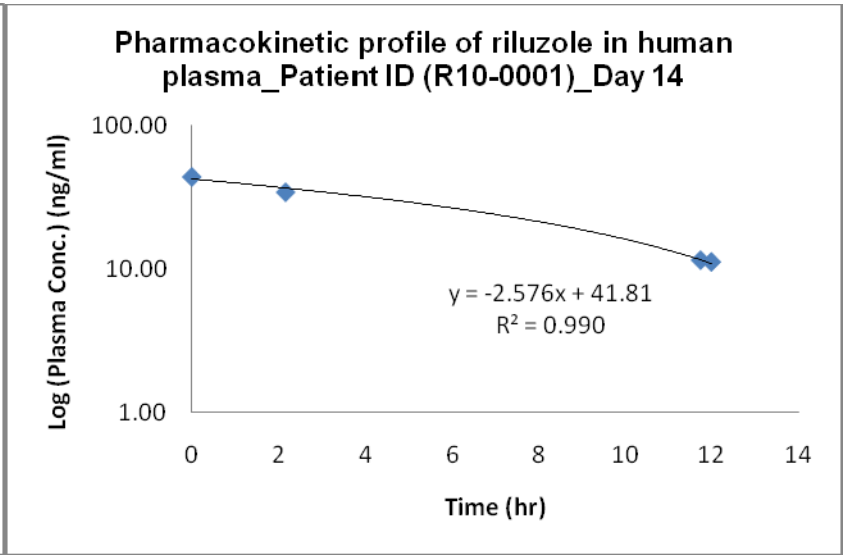
(O)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R07-0008)_Day 14
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
y = -20.98x + 260.4
R² = 0.949
0
2
4
6
8
10
12
14
Time (hr)

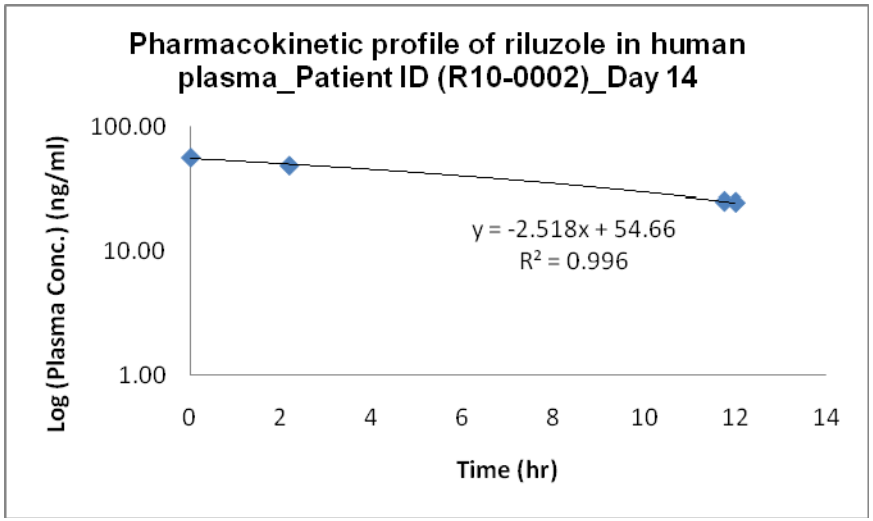
(P)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0001)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
y = -5.084x + 99.8
R² = 0.995
0
2
4
6
8
10
12
14
Time (hr)

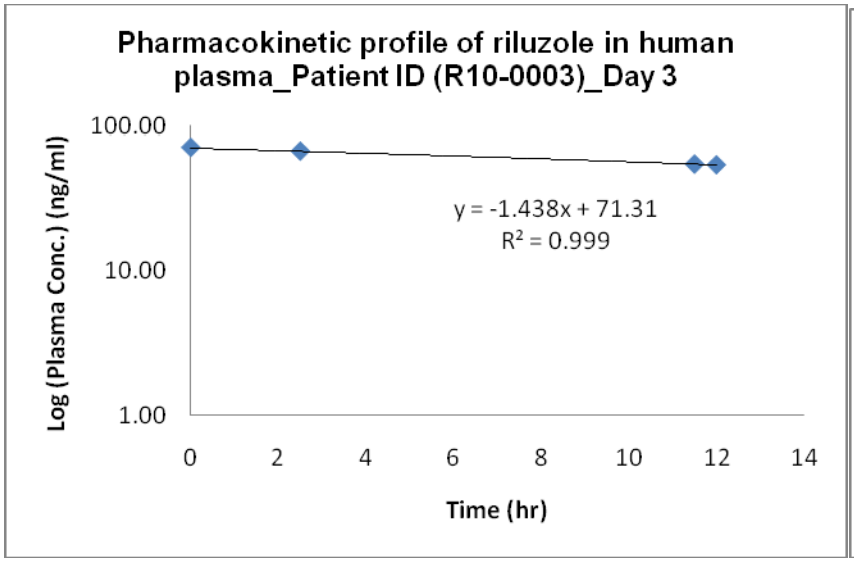
(Q)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0001)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -2.576x + 41.81
R² = 0.990
0
2
4
6
8
10
12
14
Time (hr)

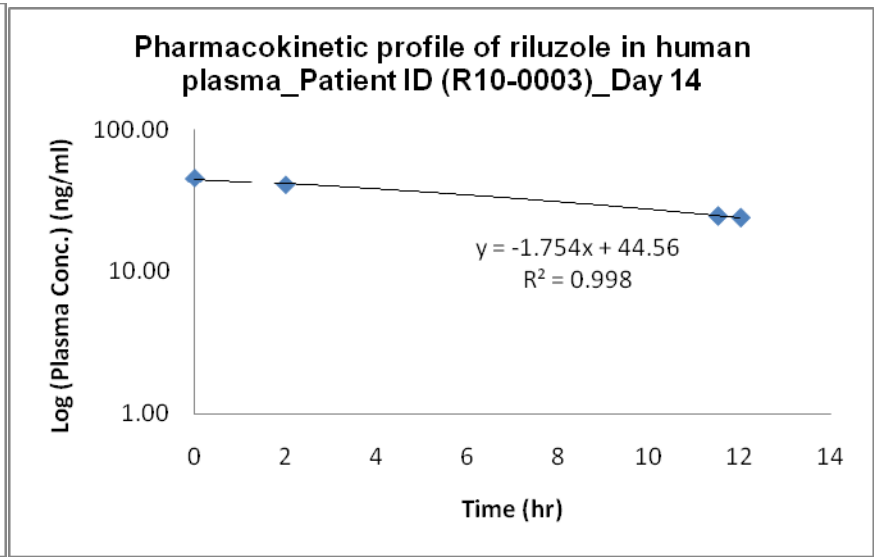
(R)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0002)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -2.518x + 54.66
R² = 0.996
0
2
4
6
8
10
12
14
Time (hr)

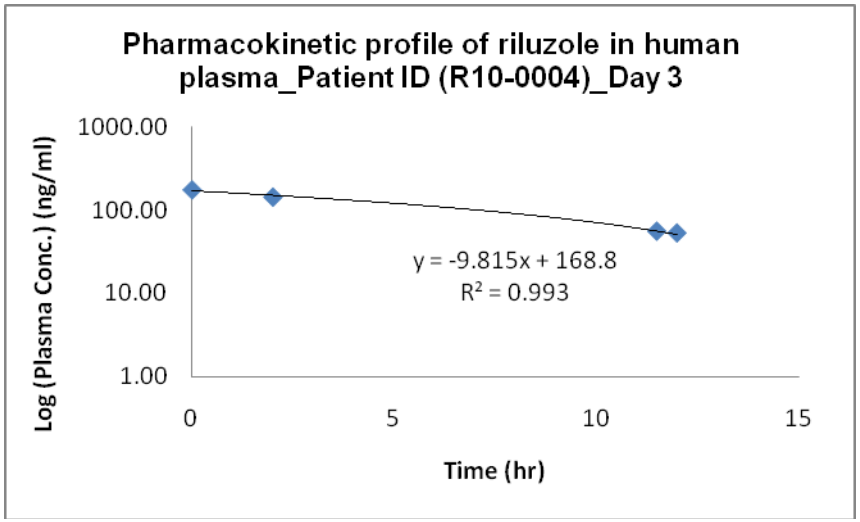
(S)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0003)_Day 3
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -1.438x + 71.31
R² = 0.999
0
2
4
6
8
10
12
14
Time (hr)

(T)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0003)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -1.754x + 44.56
R² = 0.998
0
2
4
6
8
10
12
14
Time (hr)

(U)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0004)_Day 3
Log (Plasma Conc.) (ng/ml)
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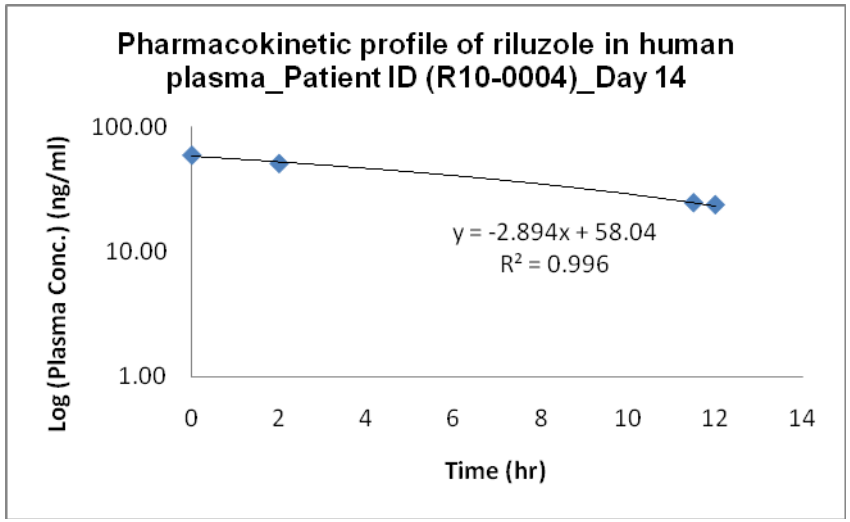
100.00
10.00
1.00
y = -9.815x + 168.8
R² = 0.993
0
5
10
15
Time (hr)

(V)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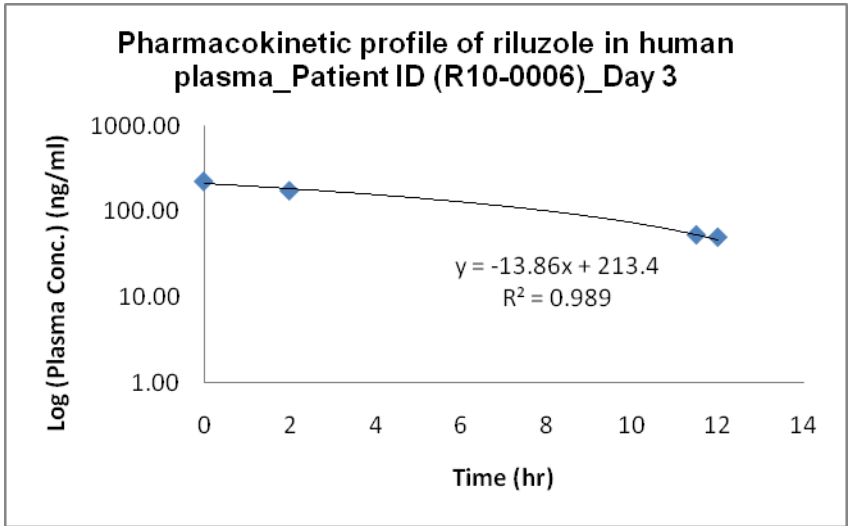
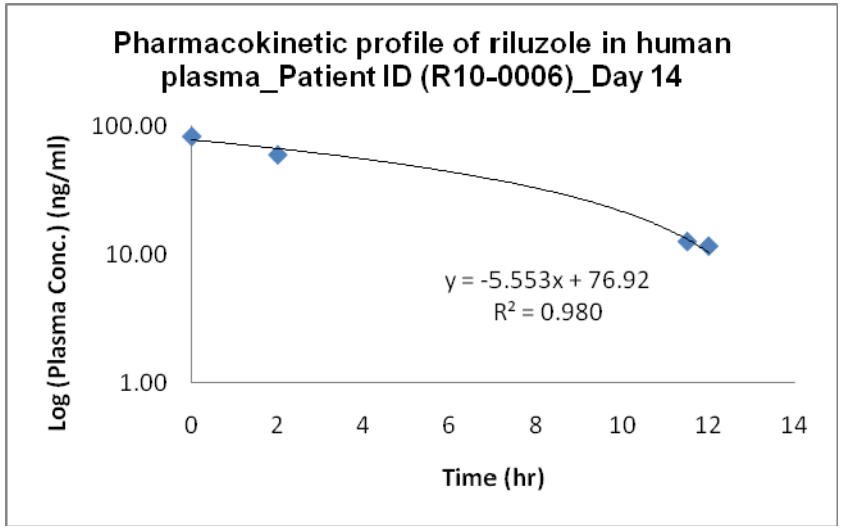
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
y = -2.894x + 58.04
R² = 0.996
0
2
4
6
8
10
12
14
Time (hr)

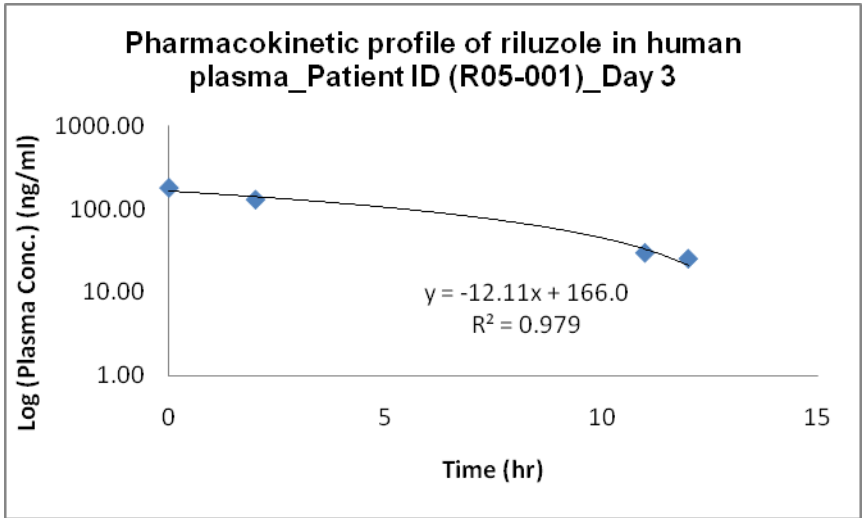
(W)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0006)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
0
2
4
6
8
10
12
14
Time (hr)
y = -13.86x + 213.4
R² = 0.989

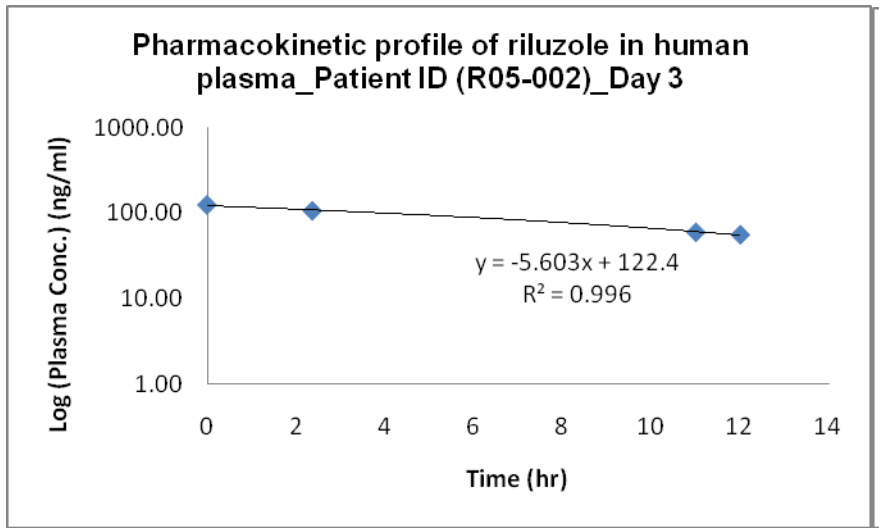
(X)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R10-0006)_Day 14
Log (Plasma Conc.) (ng/ml)
100.00
10.00
1.00
0
2
4
6
8
10
12
14
Time (hr)
y = -5.553x + 76.92
R² = 0.980

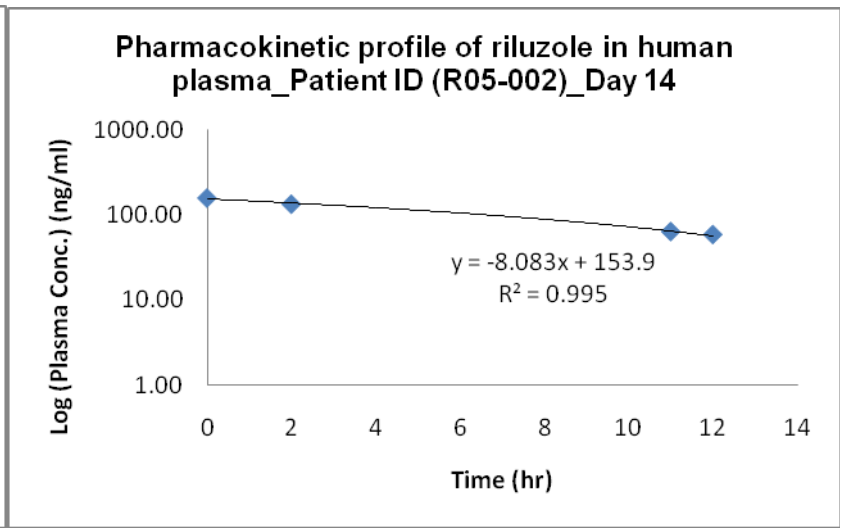
(Y)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R05-001)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
0
5
10
15
Time (hr)
y = -12.11x + 166.0
R² = 0.979

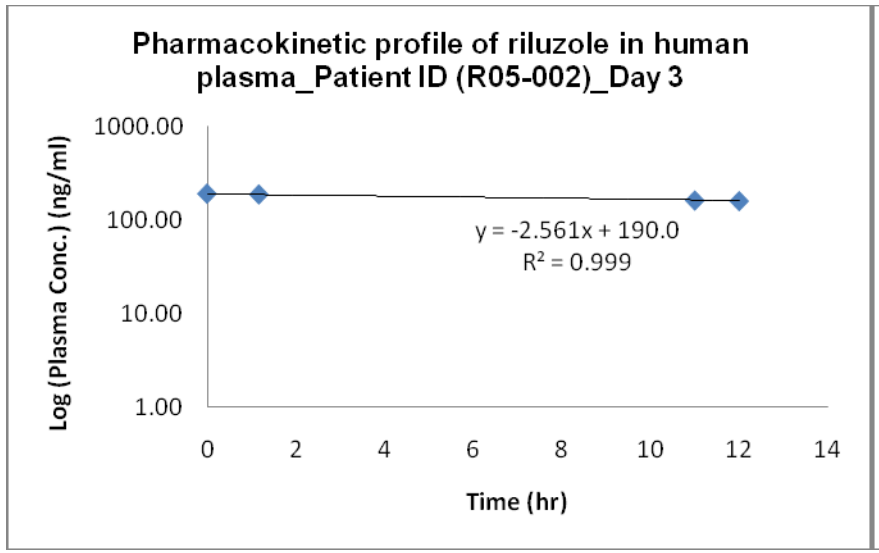
(Z)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R05-002)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
0
2
4
6
8
10
12
14
Time (hr)
y = -5.603x + 122.4
R² = 0.996

(AA)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R05-002)_Day 14
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
0
2
4
6
8
10
12
14
Time (hr)
y = -8.083x + 153.9
R² = 0.995

(AB)


Pharmacokinetic profile of riluzole in human plasma_Patient ID (R05-002)_Day 3
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
0
2
4
6
8
10
12
14
Time (hr)
y = -2.561x + 190.0
R² = 0.999

(AC)

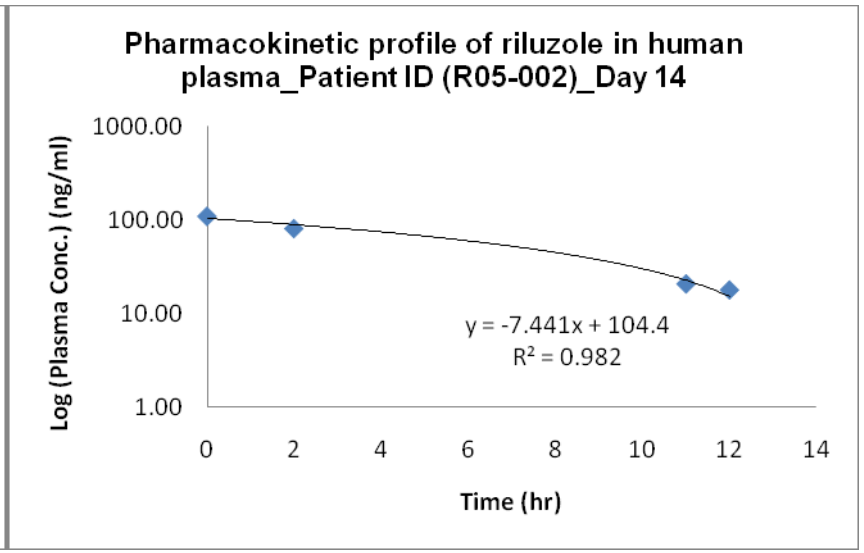

Pharmacokinetic profile of riluzole in human plasma_Patient ID (R05-002)_Day 14
Log (Plasma Conc.) (ng/ml)
1000.00
100.00
10.00
1.00
0
2
4
6
8
10
12
14
Time (hr)
y = -7.441x + 104.4
R² = 0.982

Figure 1. Pharmacokinetic Profiles of Riluzole in Plasma for Patient R07-0001 on Day 3 (Fig. A) and Day 14 (Fig. B), R07-0002 on Day 3 (Fig. C) and Day 14 (Fig. D), R07-0003 Day 3 (Fig. E) and Day 14 (Fig. F), R07-0004 Day 3 (Fig. G) and Day 14 (Fig. H), R07-0005 Day 3 (Fig. I) and Day 14 (Fig. J), R07-0006 Day 3 (Fig. K) and Day 14 (Fig. L), R07-0007 Day 3 (Fig. M), R07-0008 Day 3 (Fig. N) and Day 14 (Fig. O), R10-0001 Day 3 (Fig. P) and Day 14 (Fig. Q), R10-0002 Day 14 (Fig. R), R10-0003 Day 3 (Fig. S) and Day 14 (Fig. T), R10-0003 Day 3 (Fig. U) and Day 14 (Fig. V), R10-0006 Day 3 (Fig. W) and Day 14 (Fig. X), R05-001 Day 3 (Fig. Y), R05-002 Day 3 (Fig. Z) and Day 14 (Fig. AA) and R05-003 Day 3 (Fig. AB) and Day 14 (Fig. AC).

Table III Estimated Pharmacokinetic Parameters of Riluzole in Spinal Cord Injured Patients Enrolled in Clinical Phase I Trial

Sunject ID	$AUC_{(0-\infty)}$ (ng*hr/ml)		Half-life ($t_{1/2,}$ hr)		Elimination rate (k, hr^{-1})		Clearance (CL, L/hr)		Volume of Distribution (Vd, L)	
	Day 3	Day 14	Day 3	Day 14	Day 3	Day 14	Day 3	Day 14	Day 3	Day 14
R07-0001	1081.82	231.42	9.7	5.0	0.072	0.139	46.22	216.06	645.51	1552.16
R07-0002	1167.37	556.94	11.3	4.9	0.061	0.142	42.83	89.78	701.00	632.67
R07-0003	844.16	456.11	3.9	8.4	0.177	0.083	59.23	109.62	334.26	1325.56
R07-0004	1453.26	420.13	4.7	9.3	0.147	0.074	34.41	119.01	234.05	1603.92
R07-0005	1399.83	493.12	8.3	9.0	0.084	0.077	35.72	101.39	425.22	1321.96
R07-0006	1247.43	400.07	3.1	8.5	0.222	0.082	40.08	124.98	180.23	1520.42
R07-0007	3483.02		49.5		0.014		14.36		1025.38	
R07-0008	1415.84	1462.40	8.1	2.6	0.085	0.264	35.31	34.19	413.52	129.75
R010-0001	1311.17	405.61	8.7	6.1	0.080	0.113	38.13	123.27	479.07	1092.84
R010-0002		832.49		10.2		0.068		60.06		885.85
R010-0003	3084.62	847.02	29.9	12.9	0.023	0.054	16.21	59.03	698.68	1101.31
R010-0004	1813.13	790.48	7.0	9.1	0.100	0.076	27.58	63.25	276.60	827.92
R010-0005										

R010-0006	1902.40	562.47	5.6	4.3	0.125	0.163	26.28	88.89	210.60	547.04
R05-001	1205.13		4.3		0.162		41.49		256.90	
R05-002	1902.07	1954.78	10.5	8.4	0.066	0.082	26.29	25.58	397.09	311.17
R05-003	12940.98	792.38	47.1	4.6	0.015	0.152	3.86	63.10	262.84	415.41
Mean_all	1601.99 ± 2310.84		10.86 ± 11.49		0.104 ±0.058		91.30 ± 48.13		436.06 ± 237.44	
Mean_Day 3	2416.82 ± 3000.88		14.11 ± 15.27		0.096 ± 0.061		32.53 ± 13.93		947.71 ± 487.30	
Mean_Day 14	728.96 ± 465.42		7.38 ± 2.84		0.112 ± 0.056		60.90 ± 45.45		683.07 ± 454.03	

Appendix:

Table IV Pharmacokinetic Summary of Multiple Dose Studies of Riluzole in Twelve White Subjects (Liboux A et al., 1997)

Dose (mg)	C_{max} (ng/ml)	C_{min} (ng/ml)	T_{max} (hr)	$AUC_{(0-\infty)}$ (ng*hr/ml)	$t_{1/2\alpha}$ (hr)	$t_{1/2\beta}$ (hr)	Vd_{ss} (L)	Clearance (L/hr)
50 bid	172 (72)	21 (12)	0.8 (0.5)	654 (280)	1.5 (0.5)	14.7 (4.9)	245 (69)	47.4 (8.4)

The values of Vd_{ss} and clearance: from 30-min intravenous infusion of 50mg riluzole in 16 healthy male volunteers.

Comments: 12 healthy male volunteers were administrated 50mg riluzole twice for 10 days (day 3-13). Blood samples were collected immediately before and at 0.5, 1, 1.5, 2, 4, 6, 8, 12, 16, 24, 36 and 48 hours after the final morning dose on day 13 as well as at 1 hour after administration on days 5 and 10.

Table V Mean Pharmacokinetic Data of Riluzole in Healthy Elderly Volunteers (Liboux et al., 1999)

Subjects (n)	T_{max} (hr) (CV%)	C_{max} (ng/ml) (CV%)	$AUC_{(0-12hr)}$ (ng*hr/ml) (CV%)	CL/F (L/hr/kg) (CV%)	$t_{1/2}$ (hr) (CV%)
Elderly males (9)	0.75 (0.5-1.5)	280 ± 95 (34)	1009 ± 299 (30)	0.76 ± 0.28 (37)	42.08 ± 11.31 (27)
Elderly females (9)	0.75 (0.5-1.0)	262 ± 149 (57)	1049 ± 502 (48)	0.96 ± 0.54 (57)	38.52 ± 5.55 (14)
Elderly males +females (18)	0.75 (0.5-1.5)	271 ± 122	1029 ± 401	0.86 ± 0.43	40.30 ± 8.84
Young males (9)	0.75 (0.5-3.0)	200 ± 103 (52)	858 ± 433 (50)	0.93 ± 0.37 (40)	49.33 ± 11.08 (22)
Young females (9)	0.50 (0.5-1.0)	289 ± 163 (57)	880 ± 522 (59)	1.08 ± 0.52 (48)	48.8 ± 11.49 (24)
Young males +females (18)	0.75 (0.5-3.0)	244 ± 140	869 ± 465	1.01 ± 0.44	49.03 ± 10.93

Comments: Commencing on day 1, each subject received a daily dose of 100 mg riluzole (administered as a 50 mg film-coated tablet twice daily, one in the morning at 8 a.m. and one in the evening at 8 p.m. before meals) for 4 days and a single dose of 50 mg riluzole on the morning (8 a.m.) of day 5, administered 2 hours before breakfast. Blood samples (approximately 5 ml) were collected into lithium heparin tubes before the morning dose of riluzole on days 1 to 4 and before and at 0.5, 0.75, 1, 1.25, 1.5, 2, 3, 4, 6, 8, 12, 16, 24, 30, 36, 48, 60, and 72 hours after the last dose of riluzole on the morning of day 5.

References:

1.Le Liboux A, Lefebvre P, Le Roux Y, Truffinet P, Aubeneau M, Kirkesseli S, Montay G. Single- and multiple-dose pharmacokinetics of riluzole in white subjects. J Clin Pharmacol. 1997 Sep;37(9):820-7.

2. A Le Liboux, JP Cachia, S Kirkesseli, JY Gautier, C Guimart, G Montay, PA Peeters, E Groen, JH Jonkman and J Wemer. A Comparison of the Pharmacokinetics and Tolerability of Riluzole after Repeat Dose Administration in Healthy Elderly and Young Volunteers. J Clin Pharmacol. 1999 May;39(5):480-6.

Table 1

Principles of Treatment Selection for NACTN Clinical SCI Trials. (Dec. 2010).

Developed by the NACTN Treatment Strategy Selection Committee.

1. The agent should have shown positive results in pre-clinical trials in SCI in more than one laboratory, in more than one species, and in more than one experimental model of SCI.

2. NACTN trials can be conducted in patients with SCI in any of the phases of SCI-acute, subacute or chronic

3. Trials should be conducted utilizing treatment strategies that promote neuroprotection or regeneration or a combination of these two mechanisms.

4. Therapeutic strategies can range from simple chemicals to complex proteins, or cells, bio-engineered platforms or physical modalities and can be single strategies or combinations, one-dose, or multiple-dose.

5. The routes of administration of the strategy should be as simple as possible, but it is recognized that complex methods of administration may be required such as the intrathecal or intra-lesional route.

6. 3. Some trials should be directed towards specific complications of SCI such as post-traumatic syringomyelia.

7. Strategies that have already been tested in humans for effectiveness in another condition and proven to have acceptable toxicity are especially attractive.

8. The choice of strategy will be influenced by the likelihood of funding from government, private agency or industry.

Table 2

List of Potential Strategies for NACTN Trials with Details (Dec, 2010)

Developed by the NACTN Developed by the NACTN Treatment Strategy Selection Committee

A. Pharmacological Agents

1. **Chondroitinase ABC**. Has been positive in several trials in rodents, and in several types of experimental injury models but not in other species. No data in humans. Acorda Therapeutics, Inc.

2. **Thyrotropin Releasing Hormone (TRH).** Alan Faden has contacted NACTN about TRH. We are unaware of any new information on this agent. TRH was effective in a very small single center trial reported in 1995 in 20 patients. There is no new compelling data.

3. **Growth factors- BDNF, CNTF**. No interest at this time. Several expensive and unsuccessful trials in other conditions such as ALS, with side-effects.

4. **Cethrin.** Phase 1 trial has been reported but not published with favourable effects and minimal side-effects. Remains attractive candidate for Phase 3 trial.

5. **Anti-Nogo A** (either individual NACTN centers or as a NACTN project). Phase I trial has been completed. Route of administration has changed from continuous catheter to intermittent lumbar puncture. Still highly regarded as a candidate for a larger trial.

6. **Glyburide. (Glibenclamide).** Marc Simard would be keen to have NACTN involved and the Remedy company is involved. Preclinical trials are positive mainly, but the Popovitch NIH-sponsored replication was negative. Further pre-clinical work is necessary. Human trials in other conditions may proceed. Toxicity studies in normal humans have been completed

7. **Riluzole** (ready for Phase 3 Trial if Phase 1 trial shows no toxicity). At this point, this drug is favoured for the next NACTN trial.

8. **Asubio Agent**. Asubio Pharmaceuticals, Inc. has contacted NACTN, but no firm plan has been developed, and we do not know which of the drugs in the pipeline of this company (Daiichi-Sankyo) company is being offered for SCI.

9. **Minocycline.** Canadian multi-centre, Phase 3, trial to begin in 2011 funded by the Rick Hansen Foundation.

10. **Polyethylene Glycol/Magnesium**. No major interest at this time.

11. **MAP4343 is a pregnenelone** derivative developed in France (Baulieu). A phase 2 trial will be conducted in Paris with 60 SCI patients. This group would like NACTN's involvement. No firm plan at this time.

B. Cell-Based Therapies

1. **Human Embryonic Stem Cells** with oligo differentiation (Geron, Inc.). This trial has begun in the USA. NACTN as a group was not asked to participate

2. **Human Neural Stem Cells** (Neuralstem, Inc.) Reviewed positively at FDA. Have FDA permission to do a trial in ALS. James Harrop has interacted with this group and will continue to explore a possible SCI trial for NACTN with them.

3. **Human Fetal Brain Stem Cells** (StemCells, Inc.). Zurich trial announced (Armin Curt) in thoracic cases ASIA A-C cases 3-12 months after SCI with direct injection into the cord. NACTN was not asked to participate as a group.

4. **Human Bone Marrow Stem Cells**. The group does not think there is any established therapeutic advantage to these cells. There is no definite evidence that they can differentiate into functionally active neural cells. Major advantage is that they are autologous.

5. **Mesenchymal Stem Cells**. No firm interest at this time.

6. **Olfactory Ensheathing Cells (OECs)**. Australian small Phase1 study already reported. Alan Mckay-Sim of Australia is currently organizing a multi-centre trial, and Michael Fehlings has met with him. Jim Guest has used OECs in a primate model. Inconsistencies in type of "OEC" cell being used in various centres (eg. Huang in Beijing). Major advantage is that they are autologous. Michael Fehlings will continue to explore.

7. **Schwann Cells.** The Miami group has been working towards a clinical trial for many years, and has found positive results in rodents and primates, both species receiving autologous cells. There is some funding available from a foundation. Jim Guest has contacted Dalton Dietrich who is in favour of including NACTN in this trial. It would involve autologous nerve biopsies at 1-5 days, expansion in culture for 21-28 days and then transplantation in the subacute phase at about 30 days after injury. A Phase 1 trial in ASIA-A thoracic injuries is planned in 12 patients. Pre-IND meeting already held.

8. **Human Neural Precursor Cells**. Toronto experiments. No firm plan at this time.

9. **Human Umbilical Cord Stem Cells**. Human trial currently underway in China. Small amount of preclinical data that is not impressive.

C. Bioengineered Platforms

- **Hyaluronic Acid/Methylcellulose Medium (HAMC)**. Agent used in many pre-clinical trials in Toronto. No plans for clinical trial at this time.

D. Physical Modalities

1. **Systemic Hypothermia**. Possible NIH sponsored trial may go forward. No plan for NACTN involvement

2. **Electrical Stimulation.** No contact with NACTN from the Purdue/Indiana Richard Borgens/Scott Shapiro group that did the Phase 1 trial.

Incidence and Severity of Acute Complications after Spinal Cord Injury

Running Title: **Acute Complications after Spinal Cord Injury**

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Abstract

Knowledge of the nature and the time of occurrence of acute complications after are important in their prevention, management and prognosis of patients with spinal cord injury (SCI). The present study uses prospective consecutively collected data to determine the proportional incidence and severity of the major complications that occur in the acute phase of SCI. The cohort consists of 315 acute traumatic SCI patients drawn from nine clinical centers in North America admitted within 24 hours of injury. The study population was 79% male with a median age of 44 years. The leading causes of injury were falls (37%) and motor vehicle accidents (28%). The distribution of the American Spinal Injury Association Impairment Scale (AIS) severity of injury for patients with neurological deficit was AIS A (40%), AIS B (11%), AIS C (15%) and AIS D (29%). Forty-six percent of SCI patients had multiple mild, moderate or severe complications. Complications were significantly associated with the AIS grade; 84% of AIS A and 25% of AIS D patients had at least one complication. Seventy-eight percent of complications occurred within 14 days of injury. The most frequent types of severe and moderate complications were respiratory failure, pneumonia, anemia, cardiac dysrhythmias and bradycardia. The

mortality rate during acute care was 3.5% (95% CI: 1.8%-6.2%). Older age (p = 0.04) and pre-existing morbidity (p = 0.01) were significantly associated with mortality. The data reported can serve as a resource for evaluating the safety of new therapies for SCI.

Key words: spinal cord injury, complications, adverse events

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[6] University of Louisville, Louisville

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[8] Walter Reed Army Medical Center, Washington DC

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Introduction

Individuals who have sustained spinal cord injury (SCI) are highly susceptible to physiological disturbances and medical complications that adversely affect their neurological outcome (Aito et al., 2003). Knowledge of the nature and the time of occurrence of complications after SCI are important in their prevention. Such information can also influence the choice of new therapies for SCI. The high probability of a patient developing pneumonia within the first two weeks following SCI would constrain the use during that time of a therapy that impairs the immune system, which has been found to be depressed after SCI (Riegger et al., 2009).

The majority of reports of complications after SCI have been of chronic complications which were studied months after injury or have been of complications that were ascertained by retrospective review of patients' medical records (New and Jackson, 2010; Boakye et al., 2008; Haisma et al., 2007). The present study was designed to use prospective consecutively collected data to determine the proportional incidence and severity of the major types of medical and surgical complications that occur in the acute and sub-acute phases of SCI.

Detailed analyses of pulmonary, infectious and surgical complications, and the relationship of surgery to complications and the relationship between complications and neurological function including deterioration will be reported in subsequent papers.

Methods

Study Subjects

The study subjects were traumatic SCI patients admitted to hospitals of the North American Clinical Trials Network (NACTN), a network of departments of neurosurgery that joined together in 2004 to bring therapies for SCI from the laboratory to clinical trials. With initial funding from the Christopher Reeve Foundation in 2004, NACTN began with a core of five clinical centers and has expanded with additional support from the Department of Defense, Telemedicine and Advanced Technology Research Center (TATRC), United States Army Research and Materiel Command (USAMRMC), to nine clinical centers in 2010. Table 1 lists the clinical centers, the Data Management Center and the NACTN investigators.

Study Population

The study population includes consecutive patients admitted to a NACTN clinical center from June 25, 2005 through November 2, 2010 who met the following criteria; 1) initial AIS grade A, B, C, or D; 2) 18 years-of-age or older; 3) admitted to a NACTN clinical center within 24 hours of an initial SCI; 4) discharged from acute care prior to November 2, 2010; and 5) provided written informed consent to participate in the NACTN SCI registry. All penetrating and non-penetrating injuries were included. A total of 315 SCI registry patients met these criteria.

Patient Care

Patients received the current standard of care for SCI as described in the *Guidelines for the Management of Acute Cervical Spine and Spinal Cord Injuries* (Hadley et al., 2002). Treatment modalities included rapid ventilatory, cardiovascular and nutritional support, reduction of vertebral subluxations, surgical decompression of the spinal cord and vertebral

stabilization, prophylactic measures to prevent deep vein thrombosis using compression devices or anticoagulation using heparin or low molecular weight heparin.

NACTN SCI Registry

Patients with a traumatic SCI and neurological deficit, age 18 or older, admitted to a neurosurgical service of the NACTN hospitals were enrolled in the SCI registry after informed consent was obtained. The registry, informed consent form and consenting process were approved by the Institutional Review Board of each center and the Human Research Protection Office (HRPO), USAMRMC. Data were collected under a standardized protocol and manual of operations designed specifically for the registry. The NACTN protocol and manual of operations are available upon request. All data were submitted to the Data Management Center and were subjected to multiple manual and electronic data quality control procedures. De-identified data are stored in secure password protected computer databases, created specifically for research purposes.

All data were collected prospectively starting at the time of admission to a NACTN clinical center. The registry data includes extensive demographic information, past medical history, pre-injury medication use, circumstances of injury, time of injury, and the time of arrival to the treating NACTN hospital. Further detail was elicited about the condition of the patient on arrival and included a clinical evaluation, measurement of state of consciousness with the Glasgow Coma Scale (GCS) and of associated injuries with the Abbreviated Injury Scale.

The International Standards for Neurological Classification of Spinal Cord Injury (ISNCSCI) examination was performed and the American Spinal Injury Association Impairment Scale (AIS) was scored on admission and at key times throughout the patients' hospital and post-hospital course (Marino et al., 2003). All examiners received training on performing the examination and study procedures. Data were also collected on radiographic findings, non-

operative and operative treatments, timing of treatments, and perioperative complications. Requirements for mechanical ventilation, discharge AIS score, and the facility to which the patient was transferred were recorded in the discharge form. After acute care discharge, follow-up was scheduled at approximate intervals of 3, 6 and 12 months after discharge. The follow-up registry protocol includes: the AIS Impairment Scale, and where appropriate, the Functional Independence Measure FIM™, the Spinal Cord Independence Measure (SCIM), and the Walking Index for Spinal Cord Injury (WISCI) evaluations.

Definition and Categorization of Complications

A complication (adverse event) was defined as an event that required medical or surgical treatment and/or prolonged hospitalization. For accurate and efficient collection of data on complications it was necessary to create forms that would strike a balance between creating an excessive number of categories, a situation that would make choosing an appropriate category difficult, versus over-simplification and failure to capture the diversity of the complications that patients sustain. The following eight disease/systems were selected for recording complications: infectious, pulmonary, hematologic, cardiac, skin, gastrointestinal/genitourinary, neuropsychiatric, and death. Forms were developed for each category with a list of the types of complications that occur within that category.

Infectious complications include pneumonia, urinary tract infection, empyema, wound infection, sepsis, abscess, infectious diarrhea, and central nervous system infection including meningitis.

Pulmonary complications include acute lung injury, acute respiratory distress syndrome, respiratory failure, pulmonary embolus, pleural effusion, lobar collapse, mucus plug, pneumothorax, and hemothorax.

Hematologic complications include deep vein thrombosis, anemia, thrombocytopenia, and coagulopathy.

Cardiac complications include Bradycardia (heart rate < 50 bpm), other dysrhythmia, cardiac arrest, myocardial infarction, shock (systolic blood pressure < 80 mm Hg), congestive heart failure, and cardiogenic pulmonary edema.

Skin complications include sacral, heel, scapular, occipital, trochanteric, decubiti and operative wound problems..

Gastrointestinal/genitourinary complications include hemorrhage, ileus, pancreatitis, cholecystitis, hematuria and acute renal failure.

Neuropsychiatric complications include depression, psychosis, seizure, and cognitive deterioration.

Cause of death and contributing causes and complications were recorded. Complications that did not fall into one of the named complications were recorded by their best possible description within one of the major categories of complications.

The severity of each complication was categorized using the following guidelines*:* *Mild* if the complication had no residual effect, if there was no treatment required, or if the treatment was benign; *Moderate* if the complication had permanent adverse effect or required a more aggressive treatment; *Severe* if the complication required an aggressive treatment with a permanent impact on outcome. Surveillance for the occurrence of complications was performed by neurosurgeons and intensivists and was recorded by clinical research nurses dedicated to the study.

Statistical Methods

The study population description includes age, gender, cause and circumstance of injury, SCI injury type and level, AIS grades at admission, hospital arrival times, acute care length of stay, and discharge disposition. Incidence of complications is reported as the number and percent of patients (proportional incidence) who experienced at least one mild, moderate, or severe complication. Complications are reported by type and frequency. Descriptive statistics reported include means, standard deviations, and percentiles. Comparisons of categorical variables were computed using uncorrected exact tests and confidence intervals for binomial proportions; two-sample tests and confidence intervals for comparison of binomial proportions and Pearson chi-square tests for associations and trends in proportions. All statistical computations were performed by using Stata statistical software (StataCorp, 2009. Stata Statistical Software, Release 11.1, College Station, TX: StataCorp LP).

Results

The report is arranged in the following manner: Demographic characteristics, cause of injury, time to hospital, type and severity of injury, anatomical level, coma score and co-morbidities are reported in Table 2; aspects of medical and surgical treatment and length of stay are given in Tables 3-5; data on the types, severity and incidence of complications that occurred from admission to time of discharge, or a maximum of ninety days, are presented in Tables 6-12

Demographic and Baseline Patient Characteristics

The 315 SCI patients were predominately male (79%). The median age of patients was 44 years with an age range from 18 to 87 years. Approximately 25% were younger than 29 years of age, and 25% were older than 57 years of age. The three leading causes of injury were accidental falls (37%), motor vehicle (28%), and motorcycle/off-road vehicle (11%).

Fifty-seven percent of the 315 patients were transported directly from the scene of injury to a NACTN Center with a median arrival time of approximately 1.1 hours. Forty-three percent were transferred to a NACTN Center from an intermediate-care hospital with a median post-injury arrival time of approximately 8.2 hours.

Injuries were classified as “Blunt” (263 cases) (flexion, extension, subluxation) or “Crushing”, a subgroup of blunt injury, (36 cases) (compression, burst fracture) and “Penetrating” (13 cases).

There was a significant bimodal distribution of AIS grade severity of neurological injury with peaks at the most severe and at the milder ends of the spectrum of AIS grade severity (Pearson’s X^2, $p < 0.001$). The incidence of AIS A severity was 40%, and the incidence of AIS D severity was 29%. AIS B and C each had a significantly lower incidence, 17% and 15% respectively (Table 2).

Levels of injury of SCI were cervical (77%); thoracic (18%); lumbar/sacral (4%) and spinal cord injury without radiographic abnormality (1%).

The distribution of Glasgow Coma Scale scores shows that 11% of the patients had a mild traumatic brain injury; 8% sustained a moderate and 2% a severe brain injury.

The prevalence of one or more pre-existing medical co-morbidities among all patients was 35%. The leading reported causes of medical co-morbidities were hypertension, suspected history of alcohol or drug abuse, and diabetes.

The Abbreviated Injury Scale 90 (www.trauma.org/archive/scores/ais.html) was adopted mid-course in the registry program for the evaluation of the severity of somatic injury. Sixty-four percent of the 315 patients have Abbreviated Injury Scale 90 scores. Of these 1% had severe or critical abdominal injuries, 6% severe or critical chest injuries, and 16% severe or critical head and neck injuries.

Aspects of Medical and Surgical Treatment and Length of Stay in Acute Care

Table 3 summarizes the use of corticosteroids. About two-thirds of AIS A, AIS B, and AIS C patients received corticosteroids and 56% of AIS D patients received corticosteroids. Methylprednisolone was the corticosteroid most frequently administered.

The great majority of patients in all AIS grades underwent either anterior, posterior, or combined procedures for surgical decompression and stabilization. AIS A patients had the highest incidence of surgery, 93%; AIS B, 92%, AIS C, 91%, and AIS D 89% (Table 4). Generally, surgeries were performed early with nearly 50% performed within two days of injury, and 75% within four days of injury.

Length of stay is summarized in Table 5. Approximately one-fourth of all AIS A and B patients had acute care lengths of stay exceeding 30 days. More than half (61%) of AIS C patients had lengths of stay of 14 days or less and about half (48%) of AIS D patients had acute

lengths of stays of less than 7 days. Of 315 patients, 77% were discharged to a SCI rehabilitation unit or hospital; 13% were discharged home; 3% were discharged to a skilled nursing facility or nursing home, and 2% were continued on long-term hospital care. Discharge status was unknown for 12 of the 315 (3.8%) patients.

Incidence of Complications, Relationship to Severity and Type of Injury and to Patient Characteristics

Of the total of 315 patients, 167 (58%) developed one or more mild, moderate, or severe complication. Multiple complications were common with 144 patients (46%) developing 2 or more mild, moderate, or severe complications (Table 6).

Table 7 gives the proportional incidence of mild, moderate, or severe complications by AIS severity. The most severely injured patients had the highest incidence of complications. Among 126 AIS A patients, 106 (84.1%) developed at least one mild, moderate, or severe complication. Proportional incidence of reported complications declined significantly and linearly from 84.1% for AIS A patients to 25.3% for AIS D patients (chi-square test for trend, $p < 0.001$).

Table 8 gives the proportional incidence of complications by selected baseline characteristics of the 315 SCI patients. Age, gender, concurrent morbidity, hospital of first treatment, injury type and highest level of injury are only very weakly related to the proportional incidence of subsequent SCI complications. However, proportional incidence of complications varied significantly by injury type (Pearson X^2, $p = 0.020$). Of penetrating injuries (12 bullet, 1 knife), 84.6% had at least one mild, moderate or severe complication; 58.9% of blunt injuries and 41.7% of crushing injuries had at least one mild, moderate or severe complications.

Frequency of Occurrence of Specific Complications

Table 9 gives the frequency distribution of 845 acute and subacute complications experienced by the 315 patients within 90 days of SCI. The leading categories of complications are pulmonary, infectious, hematologic, and cardiac, accounting for 76% of all 845 incident complications. Approximately 55% of all complications occurred within the first seven days following injury and 78% occurred within 14 days of injury.

Table 9 also illustrates the extent of the clustering of complications within patients. For example, 121 patients accounted for the 228 pulmonary complications whereas only 57 patients accounted for the 65 neuropsychiatric complications (primarily depression). Table 9 also gives the proportional incidence for each type of complication. Proportional incidence separates the complications into three distinct groups: pulmonary and infectious; hematologic and cardiac: and GI/GU, skin, and neuropsychiatric complications in decreasing order of frequency.

Pneumonia can be classified as either an infectious or as a pulmonary complication. Of the 315 patients, 62 patients accounted for the 74 cases of pneumonia. The incidence rate of at least one mild, moderate or severe case of pneumonia was 62/315 or 19.7%. If pneumonia is counted as a pulmonary complication, then the incidence rate for pulmonary complications increases from 38.4% to 41.6%. If pneumonia was removed from infections then the incidence rate for infections would decrease from 34.9% to 69/315 or 21.9%.

Deep vein thrombosis (DVT) was classified as a hematologic complication. DVT could also be classified as a vascular complication, a category that was not selected as one of the major headings of complications. There were 13 patients who experienced a mild or moderate DVT. No severe episodes of DVT were reported. However, two cases of pulmonary embolus were reported (Table 12). The incidence rate of deep vein thrombosis was 13/315 or 4.1%.

Distribution of Mild, Moderate and Severe Complications

Of the total number of 845 acute and subacute complications, 153 (18%) were classified as mild complications (Table 9). The four leading categories of mild complications were 1) pulmonary, 2) infectious, 3) hematologic, and 4) cardiac. These four complications accounted for 71% of the total number of the 153 mild complications reported.

Tables 10-12 give the frequency of the most common types of complications graded by severity. Note that only the leading types of complications are given within each of the seven major categories of complications.

Mild Complications (Table 10)

Pleural effusion, Acute Respiratory Distress Syndrome or Acute Lung Injury, and Respiratory Failure accounted for two-thirds of all mild pulmonary complications.

Urinary Tract Infection, Pneumonia, and Empyema accounted for 80% of all mild infectious complications.

Anemia, Coagulopathy, and Deep Vein Thrombosis accounted for 77% of all mild hematologic complications.

Cardiac arrhythmias, Shock (SBP <80 mm Hg), and Cardiogenic Pulmonary edema accounted for 58% of all mild cardiac complications.

Moderate Complications (Table 11)

Of the total number of 845 acute and subacute complications, 597 (71%) were classified as moderate complications. Similar to mild complications, the leading categories of moderate complications were pulmonary, infectious, hematologic, and cardiac. These four categories accounted for 77% of all moderate complications.

Respiratory Failure, Pleural Effusion, Lobar Collapse, and Pneumothorax accounted for two-thirds of all moderate pulmonary complications.

Pneumonia, Urinary Tract Infections, Empyema, and Wound Infection accounted for 87% of all moderate infectious complications.

Anemia, Coagulopathy, Thrombocytopenia, and Deep Vein Thrombosis accounted for 83% of all moderate hematologic complications.

Cardiac arrhythmias, Shock, and Cardiac Arrest accounted for (73%) of all moderate cardiac complications.

Severe Complications (Table 12)

Of the total number of 845 acute and subacute complications, 95 (11.2%) were classified as severe complications. The leading categories of severe complications were pulmonary, cardiac, infectious, and gastrointestinal. These four categories accounted for 81% of the 95 severe complications.

Pulmonary complications were the leading cause of severe complications accounting for 42% of all severe complications. The leading severe pulmonary complication was respiratory failure (60.0%) followed by pleural effusion (17.5%), ALI/ARDS (12.5%), and pulmonary embolus (5.0%).

Cardiac complications were the second-ranking cause of severe complications accounting for 21% of all severe complications. Sixteen patients accounted for the 20 severe cardiac complications yielding an incidence rate of 5.1% for severe cardiac complications. Bradycardia (45.0%), cardiac arrest (25.0%), and shock (20.0%) were the leading causes of severe cardiac complications.

Pneumonia and Wound Infection accounted for 79% of all severe infectious complications. Pneumonia accounted for 9 of 14 (64.3%) of severe complications due to infection.

Ileus and Acute Renal Failure accounted for 80% of all severe Gastrointestinal/Genitourinary complications.

Across all levels of severity, abruptly occurring and potentially lethal complications of pulmonary embolus (n = 2 cases), deep vein thrombosis (n = 13 cases) and gastrointestinal hemorrhage (n = 6 cases), had comparatively low incidence but are among the most serious adverse events.

Deaths

Eleven patients died during acute care or 3.5% in the total series of 315 patients. All eleven deaths (6 male and 5 female) were cervical AIS A injuries. The mortality rate among the 126 AIS A patients was 8.7%. Fatal cases were between 22 and 81 years-of-age with a mean age of 55 years. All eleven fatal cases had concurrent morbidities or significant risk factors. These included hypertension, diabetes, hepatitis C, drug or alcohol abuse, and smoking history.

Compared to non-fatal SCI cases, age and prevalence of concurrent morbidities were significantly associated with SCI death. On average, patients who died during acute care were 11 years older than non-fatal SCI cases. The mean age difference between fatal and non-fatal cases was significant (student's t-test, two-sided p-value = 0.038). The prevalence difference of concurrent morbidity between fatal and non-fatal cases was also significant (Fisher Exact test, two-sided p-value = 0.01).

Traumatic brain injury was identified as a contributing cause of death in four of the eleven deaths. Cardiopulmonary and cardiac events were contributing causes in the other seven deaths. All patients who died suffered multiple moderate and severe complications. Four of the eleven patients died within one week of their traumatic spinal cord injury; three within two weeks and four deaths occurred between 20 and 49 days after injury.

Discussion

Representative Nature of the Study Population

Contemporary prospectively collected data of the spectrum, incidence and severity of complications occurring acutely after SCI has not, to the best of our knowledge, been previously reported in the literature. Tator et al., reported on the complications and cost of management of acute SCI for 191 of the first 220 consecutive patients managed in a regional, multidisciplinary acute spinal cord injury unit from 1974 to 1981. Respiratory, gastrointestinal, thromboembolic, genitourinary complications and decubitus ulceration were associated with increases in length of stay and cost of stay.

How representative of the North American acute SCI population are the patients in the present study? All disease registries are subject to selection bias due to the demographic characteristics of the population from which subjects are drawn and as a function of the enrollment process. Seven of the nine NACTN hospitals are centrally located in major metropolitan areas from which they receive patients. The University of Maryland/Shock Trauma Hospital also receives patients from a state-wide network. The University of Virginia is in a medium-size city and also receives patients from throughout the state. With respect to the enrollment process, informed consent requirements excluded patients with who could not give consent and thereby prevented some patients with very severe associated injuries from inclusion in the registry. Therefore, the complication and death rates reported may be too low to fully represent the population of all SCI cases admitted within 24 hours of injury.

There is no absolute benchmark to evaluate whether our series is completely representative of the North American SCI population. The cohort is similar to U.S. and international literature in male to female ratios, causes of injury, mean age of patients and dominance of cervical injuries (Ackery et al., 2004; Furlan et al., (2009); National Spinal Cord Injury Statistical Center (NSCISC), 2009. NSCISC reported that in 2009, the average age at injury was 40.2 years, 81% were male, and motor vehicle crashes and falls accounted for 69% of all cases. Couris et al. (2010) reported population-based estimates of the incidence of traumatic SCI in adults living in Ontario, Canada from 2003 to 2007. In the Ontario study, the average age at injury was 51.3 years; 74% were male; motor vehicle crashes and falls accounted for 74% of all cases, and 66% were cervical injuries. These data are quite similar to the demographics and causes of injury of SCI patients reported in the current NACTN series.

The cohort is similar to some reports in the literature with respect to the death rate. The death rate during acute care was 3.5% in our series. In an analysis of 31,381 admissions of acute SCI patients who underwent spinal decompression with laminectomy and/or fusion in the U.S. and reported in the National Inpatient Sample (NIS) from 1993 to 2002, the overall mortality rate was estimated at 3.0% (Boakye et al., 2007). Furlan et al. (2009) reported on risk factors for mortality after acute traumatic SCI in a case series of 297 patients admitted to the Toronto Western Hospital from January 1996 to December 2007. The in-hospital mortality rate was 5.7% for patients with ages from 15 to 96 years (mean age 52 years, AIS A 23%, cervical injury 69%). Older age, pre-existing medical conditions and motor complete SCI were identified as significant major risk factors for in-hospital mortality. Our case series had similar results. However, a 3-fold higher death rate than in the present series was reported in the Ontario, Canada population-based series of 936 traumatic SCI, where the in-hospital death rate was 11.6% (Couris et al., 2010).

Incidence and Spectrum of Complications

The incidence of complications in the present study is higher than reported in a retrospective study by Furlan et al. (2005) who reported secondary complication within two months in a consecutive case series of 55 acute SCI patients, 38 males, and 17 females, ages 17-89 years, admitted to the Toronto Western Hospital from January 1998 to December 2000. During hospitalization 44.7% of men and 52.9% of women developed complications within two months of injury. In the present series, 57.8% of men and 57.6% of women developed a complication within 90 days.

Previous studies of complications after SCI have tended to focus on particular types of complication, particularly deep venous thrombosis (Aito et al., 2002, Ploumis et al., 2009), respiratory complications (Jackson et al., 1994), and cardiovascular complications (Furlan et al., 2008). Aito et al. (2003) analyzed complications during the acute phase of traumatic spinal cord lesions in 558 subjects, who were admitted either to a spinal unit (7 centers, about 240 patients), a rehabilitation center (25 centers) or a regional service (5 centers). The population included both traumatic and non-traumatic SCI and first-time admissions up to 60 days post- injury.
The spinal units were able to admit patients from the time of injury, and in this respect were similar to the NACTN centers. The spinal units were also the rehabilitation facility for these patients. In the spinal units the following complications were reported *on admission:* trophic skin changes 15.5%; heterotopic ossification 6.9%; urinary 2.1%; respiratory 9.4%; deep vein thrombosis 1.3%, and one case of pulmonary embolism.

Respiratory (pulmonary) complications have been recognized as the leading cause of morbidity following traumatic SCI (Jackson et al., 1994, Cardozo (2007). In the NACTN series, the incidence of either a pulmonary complication or pneumonia was 41.6%, making respiratory complications the leading cause of complications.

Cardiac complications were the second-ranking cause of severe complications in our series, accounting for 21% of all severe complications. Bradycardia, cardiac arrest, and shock were the leading causes of all severe cardiac complications whereas bradycardia, dysrhythmias and shock were the leading causes of mild and moderate complications.

The spectrum of complications and their incidence reported here may reflect three aspects of the current management of SCI which have evolved from the methods that were in place at the time of writing of many of the previous reports:(1) a greater use of surgical decompression and vertebral stabilization, reaching 93% for ASIA A patients in the present series; (2) management of SCI in critical care units with intensive physiological monitoring and a standardized protocol that facilitates detection and recording of adverse events; (3) improved survival of patients with associated critical injuries who are, nevertheless, vulnerable to infection and other complications.

Relevance of Data on Complications to Management of SCI and Development of Clinical Trials

Prospective data on complications are important for developing improved management of SCI, and emphasize the need for early recognition of respiratory failure, infection, pneumonia, anemia, coagulopathy, cardiac arrhythmias and shock and for devising better management for these physiological disturbances.

The data are also of importance in designing therapy and in planning clinical trials. The high rate of infection sustained by SCI patients after SCI would indicate that therapies that reduce the immune response should be used with great caution in the acute phase after SCI. Similarly, therapies that can cause pulmonary, cardiac and hematological complications must be carefully monitored.

The data are also important for the interpretation of safety data in phase 1 trials. Phase 1 trials involve small numbers of patients. In reports of such trials, the statement is sometimes

made that complications were observed during the course of treatment but the complications were not related to the treatment. The figurative meaning of such a statement is that the treatment being given is known from its use in other conditions, or in normal subjects, to produce certain adverse effects, and that these effects were not observed or did not occur in inordinate numbers. However, it cannot be said that the therapy did not produce an increase in complications unless the study had a control group in which the incidence of complications was prospectively recorded. Although a therapy has not been recognized to produce adverse effects in various diseases or in healthy patients it may produce unexpected complications in SCI patients. We believe that the data in the present study can serve as a benchmark for evaluating the safety of new therapies in SCI and that such data is essential for evaluation of therapy.

Future papers will address the questions of the relationship of concurrent morbidities and other risk factors on the development and time-course of specific complications, the influence of complications on neurological outcome, and the relationship between surgical procedures, complications and neurologic outcomes.

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Author Disclosure Statement

No competing financial interests exist.

Table 1. NACTN Centers and Investigators

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Table 2. Characteristics of 315 SCI cases (06/25/2005 – 11/02/2010)

Characteristic	N = 315 (%)
Gender	
Male	**249 (79%)**
Female	**66 (21%)**
Age (years)	
Mean (s.d.)	**44.6 (17.1)**
25th Percentile	**29 yrs**
Median Age	**44 yrs**
75th Percentile	**57 yrs**
Cause of Injury	
Fall	**115 (37%)**
Motor Vehicle Collision	**89 (28%)**
Motorcycle or Off-Road Vehicle	**34 (11%)**
Sport Diving	**25 (8%)**
Other Sports/Recreation/Pedestrian	**19 (6%)**
Assault	**19 (6%)**
Not Reported	**14 (4%)**
NACTN Center Admission	
Direct from Scene	**178 (57%)**
Intermediate Hospital	**137 (43%)**
Injury Type	
Blunt	**263 (84%)**
Crushing	**36 (11%)**
Penetrating	**13 (4%)**
Not Reported	**3 (1%)**
AIS Injury Severity	
A	**126 (40%)**

B	**52 (17%)**
C	**46 (15%)**
D	**91 (29%)**
Highest Level of Injury	
Cervical	**244 (77%)**
Thoracic	**56 (18%)**
Lumbar/Sacral	**13 (4%)**
SCI Without Radiographic Abnormality	**2 (1%)**
Glasgow Coma Scale at Admission	
15 (normal)	**241 (79%)**
13 to 14 (mild brain injury)	**36 (11%)**
9 to 12 (moderate brain injury)	**24 (8%)**
3 to 8 (severe brain injury)	**11 (2%)**
Not Reported	**3 (1%)**
Concurrent Pre-Injury Morbidities	
None	**206 (65%)**
Present	**109 (35%)**

Table 3. Frequency of corticosteroid use within AIS Severity Grades

Steroid Use[1]	**AIS A**	**AIS B**	**AIS C**	**AIS D**	**Total**
Yes	**85**	**35**	**29**	**51**	**200**
No	**40**	**16**	**17**	**40**	**113**
Total	**125**	**51**	**46**	**91**	**313[1]**
Percent Yes	**68.0%**	**68.6%**	**63.0%**	**56.0%**	**63.9%**

[1]Two cases (AIS A and AIS B) where steroid use was unknown

Table 4. Percent distribution of anterior and posterior surgeries within AIS Severity Grades for 315 incident SCI cases

AIS Grade	%Anterior	%Posterior	%Both	%None	%Surgery
A N = 126	12.7%	48.4%	31.8%	7.1%	92.9%
B N = 52	25.0%	46.2%	21.2%	7.7%	92.3%
C N = 46	26.1%	41.3%	23.9%	8.7%	91.3%
D N = 91	41.8%	30.8%	16.5%	11.0%	89.0%

Table 5. Percent[1] distribution of acute care length-of-stay[2] by AIS grade for 315 incident SCI cases

AIS Grade	< 7 days	8-14 days	15-21 days	22-30 days	> 30 days
A N = 125	8.8%	27.2%	20.8%	16.8%	26.4%
B N = 52	15.4%	23.1%	25.0%	13.5%	23.1%
C N = 46	21.7%	39.1%	23.9%	8.7%	6.5%
D N = 91	48.4%	29.7%	11.0%	6.6%	4.4%

[1]Percents may not add to 100% due to rounding

[2]Length-of-stay not reported for 1 case

Table 6. Distribution of mild, moderate, or severe complications for 315 SCI cases

Number of Complications	Number of Patients	Percent of Total
None	133	42.2%
1	38	12.1%
2	23	7.3%
3	27	8.6%
4 or more	94	29.8%
Total	315	100.0%

Table 7. Proportional incidence of all complications by AIS severity

AIS Severity	Patients	Complications[1]	Percent	95% Confidence Interval[2]
A	126	106	84.1%	(76.6%, 90.0%)
B	52	32	61.5%	(47.0%, 74.7%)
C	46	21	45.7%	(30.9%, 61.0%)
D	91	23	25.3%	(16.7%, 35.5%)
Total	315	182	57.8%	(52.1%, 63.3%)

[1]Number of patients with mild, moderate, or severe complications

[2]Exact Binomial Confidence Interval

Table 8. Distribution of complications by baseline characteristics

Characteristic	No complication	1 or more complications	P-value
Age (years)			
Mean (s.d.)	35.4 (15.5)	40.8 (17.5)	0.198
Gender			
Male (n = 249)	42.2%	57.8%	
Female (n = 66)	42.4%	57.6%	0.970
Pre-Existing Morbidity			
None (n = 109)	45.0%	55.1%	
Present (n = 206)	40.8%	59.2%	0.475
Registry Hospital Admission			
Direct from Scene (n = 178)	39.3%	60.7%	
Intermediate Hospital (n = 137)	46.0%	54.0%	0.235
Highest Level of Injury			
Cervical (n = 244)	41.0%	59.0%	
Thoracic (n = 56)	41.1%	58.9%	

Lumbar/Sacral (n = 13)	**61.5%**	**38.5%**	**0.340**
Injury Type			
Blunt (n = 263)	**41.1%**	**58.9%**	
Crushing (n = 36)	**58.3%**	**41.7%**	
Penetrating (n = 13)	**15.4%**	**84.6%**	**0.020**

Table 9. Distribution of complications by severity

Complication	Mild	Moderate	Severe	Total	Patients[1]	Incidence Rate
Pulmonary	**38**	**150**	**40**	**228**	**121**	**38.4%**
Infectious[2]	**30**	**131**	**14**	**175**	**110**	**34.9%**
Hematologic[3]	**22**	**105**	**3**	**130**	**72**	**22.9%**
Cardiac	**19**	**71**	**20**	**110**	**75**	**23.8%**
GI/GU[4]	**12**	**49**	**10**	**71**	**52**	**16.5%**
Skin	**17**	**45**	**4**	**66**	**48**	**15.2%**
Psychiatric	**15**	**46**	**4**	**65**	**57**	**18.1%**
Total	**153**	**597**	**95**	**845**		
%Total	**18.1%**	**70.7%**	**11.2%**	**100%**		

[1] Column gives number of patients accounting for total number of complications by type

[2] Infection includes 74 cases of pneumonia classified as mild (7), moderate (58) and severe (9) reported for 62 patients

[3]Hematologic complications include 13 DVT complications classified as mild (3) and moderate (10)

[4] GI/GU; Gastrointestinal or Genitourinary complications

Table 10. Leading causes of 153 mild complications by complication type

Complication Type	Total	Complication Cause	Number (%)
Pulmonary	**38**	**Pleural Effusion**	**18 (47.4%)**
		ALI/ARDS[1]	**4 (10.5%)**
		Respiratory Failure	**3 (7.9%)**
Infectious	**30**	**Urinary Tract Infection**	**13 (43.3%)**
		Pneumonia	**7 (23.3%)**
		Empyema	**4 (13.3%)**
Hematologic	**22**	**Anemia[2]**	**10 (45.5%)**
		Coagulopathy	**4 (18.2%)**
		Deep Vein Thrombosis	**3 (13.6%)**
Cardiac	**19**	**Bradycardia/Dysrhythmia**	**8 (42.1%)**
		Shock (SBP < 80 mm Hg)	**2 (10.5%)**
		Cardiogenic Pulmonary Edema	**1 (5.3%)**
Psychiatric	**15**	**Depression**	**8 (53.3%)**
		Cognitive Deterioration	**2 (13.3%)**
Skin	**17**	**Sacral**	**6 (35.3%)**
		Heel/Scapular/operative wound	**4 (23.5%)**
GI/GU[3]	**12**	**Ileus/Hematuria**	**3 (25.0%)**
		Cholecystitis/Pancreatitis	**2 (16.7%)**

[1] Acute Lung Injury or Acute Respiratory Distress Syndrome

[2] Anemia is defined as a hematocrit ≤ 24% or hemoglobin concentration ≤ 8.0 mg/dL for any duration or for any reason

[3] Gastrointestinal or Genitourinary complications

Table 11. Leading causes of 597 moderate complications by complication type

Complication Type	Total	Complication Cause	Number (%)
Pulmonary	**150**	**Respiratory Failure**	**52 (34.7%)**
		Pleural Effusion	**22 (14.7%)**
		Lobar Collapse	**15 (10.0%)**
		Pneumothorax	**11 (7.3%)**
Infectious	**131**	**Pneumonia**	**58 (44.3%)**
		Urinary Tract Infection	**27 (20.6%)**
		Empyema	**17 (13.0%)**
		Wound Infection	**12 (9.2%)**
Hematologic	**105**	**Anemia[1]**	**48 (45.7%)**
		Coagulopathy	**16 (15.2%)**
		Thrombocytopenia	**13 (12.4%)**
		Deep Vein Thrombosis	**10 (9.5%)**
Cardiac	**71**	**Bradycardia/Dysrhythmia**	**29 (40.8%)**
		Shock (SBP < 80 mm Hg)	**20 (28.2%)**
		Cardiac Arrest	**3 (4.2%)**
GI/GU	**49**	**Ileus/Hematuria**	**7 (14.3%)**
		Gastrointestinal Hemorrhage	**6 (12.2%)**
		Renal Dysfunction	**3 (6.1%)**
Psychiatric	**46**	**Depression**	**33 (71.7%)**
		Other[2]	**5 (10.9%)**
Skin	**45**	**Sacral**	**17 (37.7%)**
		Other Trophic[3]	**7 (15.6%)**

[1] Anemia is defined as a hematocrit ≤ 24% or hemoglobin concentration ≤ 8.0 g/dL

[2] Psychiatric Other category includes cognitive deterioration (3), psychosis (1), and seizure (1)

[3] Skin Other Trophic category includes heel (2), operative wound (2), occipital (1), scapular (1), and trochanter (1)

Table 12. Leading causes of 95 severe complications by type

Complication Type	Total	Complication Cause	Number (%)
Pulmonary	**40**	**Respiratory Failure**	**24 (60.0%)**
		Pleural Effusion	**7 (17.5%)**
		ALI/ARDS	**5 (12.5%)**
		Pulmonary Embolus	**2 (5.0%)**
Cardiac	**20**	**Bradycardia**	**9 (45.0%)**
		Cardiac Arrest	**5 (25.0%)**
		Shock	**4 (20.0%)**
		Myocardial Infarction	**1 (5.0%)**
Infectious	**14**	**Pneumonia**	**9 (64.3%)**
		Wound Infection	**2 (14.3%)**
GI/GU	**10**	**Ileus**	**4 (40.0%)**
		Acute Renal Failure	**4 (40.0%)**
Skin	**4**	**Sacral**	**3 (75.0%)**
		Operative Wound	**1 (25.0%)**
Psychiatric	**4**	**Cognitive Deterioration**	**2 (60.0%)**
		Seizure	**2 (40.0%)**
Hematologic	**3**	**Anemia**	**1 (33.3%)**
		Thrombocytopenia	**1 (33.3%)**

Table 1. NACTN Centers and Investigators

NACTN Centers	Investigators
The Methodist Hospital **Department of Neurosurgery** **Houston, TX**	**Robert G. Grossman, MD**
Thomas Jefferson University **Department of Neurological Surgery** **Philadelphia, PA**	**James S. Harrop, MD**
University of Louisville **Department of Neurological Surgery** **Louisville, KY**	**Jonathan E. Hodes, MD** **Susan K. Harkema, PhD**
University of Maryland **Shock Trauma Center** **Baltimore, MD**	**Bizhan Aarabi, MD**
University of Miami **Department of Neurological Surgery** **Miami, FL**	**James D. Guest, MD, PhD**
University of Texas Medical School at Houston **Department of Neurosurgery** **Houston, TX**	**John W. Crommett, MD**[1] **Michele M. Johnson, MD**
University of Toronto **Department of Neurosurgery** **Toronto, Ontario, Canada**	**Michael G. Fehlings, MD, PhD** **Charles Tator, MD, PhD**
University of Virginia **Department of Neurological Surgery** **Charlottesville, VA**	**Christopher I. Shaffrey, MD**
Walter Reed Army Medical Center **Department of Neurosurgery** **Washington, D.C.**	**Michael D. Rosner, MD**
University of Texas School of Public Health **NACTN Data Management Center** **Houston, TX**	**Ralph F. Frankowski, PhD** **Keith D. Burau, PhD**

[1] **Department of Critical Care, University of Texas MD Anderson Cancer Center**

Table 2. Characteristics of 315 SCI cases (06/25/2005 – 11/02/2010)

Characteristic	N = 315 (%)
Gender	
Male	**249 (79%)**
Female	**66 (21%)**
Age (years)	
Mean (s.d.)	**44.6 (17.1)**
25th Percentile	**29 yrs**
Median Age	**44 yrs**
75th Percentile	**57 yrs**
Cause of Injury	
Fall	**115 (37%)**
Motor Vehicle Collision	**89 (28%)**
Motorcycle or Off-Road Vehicle	**34 (11%)**
Sport Diving	**25 (8%)**
Other Sports/Recreation/Pedestrian	**19 (6%)**
Assault	**19 (6%)**
Not Reported	**14 (4%)**
NACTN Center Admission	
Direct from Scene	**178 (57%)**
Intermediate Hospital	**137 (43%)**
Injury Type	
Blunt	**263 (84%)**
Crushing	**36 (11%)**
Penetrating	**13 (4%)**
Not Reported	**3 (1%)**
AIS Injury Severity	
A	**126 (40%)**

B	**52 (17%)**
C	**46 (15%)**
D	**91 (29%)**
Highest Level of Injury	
Cervical	**244 (77%)**
Thoracic	**56 (18%)**
Lumbar/Sacral	**13 (4%)**
SCI Without Radiographic Abnormality	**2 (1%)**
Glasgow Coma Scale at Admission	
15 (normal)	**241 (79%)**
13 to 14 (mild brain injury)	**36 (11%)**
9 to 12 (moderate brain injury)	**24 (8%)**
3 to 8 (severe brain injury)	**11 (2%)**
Not Reported	**3 (1%)**
Concurrent Pre-Injury Morbidities	
None	**206 (65%)**
Present	**109 (35%)**

Table 3. Frequency of corticosteroid use within AIS Severity Grades

Steroid Use[1]	**AIS A**	**AIS B**	**AIS C**	**AIS D**	**Total**
Yes	**85**	**35**	**29**	**51**	**200**
No	**40**	**16**	**17**	**40**	**113**
Total	**125**	**51**	**46**	**91**	**313[1]**
Percent Yes	**68.0%**	**68.6%**	**63.0%**	**56.0%**	**63.9%**

[1]Two cases (AIS A and AIS B) where steroid use was unknown

Table 4. Percent distribution of anterior and posterior surgeries within AIS Severity Grades for 315 incident SCI cases

AIS Grade	%Anterior	%Posterior	%Both	%None	%Surgery
A N = 126	12.7%	48.4%	31.8%	7.1%	92.9%
B N = 52	25.0%	46.2%	21.2%	7.7%	92.3%
C N = 46	26.1%	41.3%	23.9%	8.7%	91.3%
D N = 91	41.8%	30.8%	16.5%	11.0%	89.0%

Table 5. Percent[1] distribution of acute care length-of-stay[2] by AIS grade for 315 incident SCI cases

AIS Grade	< 7 days	8-14 days	15-21 days	22-30 days	> 30 days
A N = 125	8.8%	27.2%	20.8%	16.8%	26.4%
B N = 52	15.4%	23.1%	25.0%	13.5%	23.1%
C N = 46	21.7%	39.1%	23.9%	8.7%	6.5%
D N = 91	48.4%	29.7%	11.0%	6.6%	4.4%

[1]Percents may not add to 100% due to rounding

[2]Length-of-stay not reported for 1 case

Table 6. Distribution of mild, moderate, or severe complications for 315 SCI cases

Number of Complications	Number of Patients	Percent of Total
None	133	42.2%
1	38	12.1%
2	23	7.3%
3	27	8.6%
4 or more	94	29.8%
Total	315	100.0%

Table 7. Proportional incidence of all complications by AIS severity

AIS Severity	Patients	Complications[1]	Percent	95% Confidence Interval[2]
A	126	106	84.1%	(76.6%, 90.0%)
B	52	32	61.5%	(47.0%, 74.7%)
C	46	21	45.7%	(30.9%, 61.0%)
D	91	23	25.3%	(16.7%, 35.5%)
Total	315	182	57.8%	(52.1%, 63.3%)

[1]Number of patients with mild, moderate, or severe complications

[2]Exact Binomial Confidence Interval

Table 8. Distribution of complications by baseline characteristics

Characteristic	No complication	1 or more complications	P-value
Age (years)			
Mean (s.d.)	35.4 (15.5)	40.8 (17.5)	0.198
Gender			
Male (n = 249)	42.2%	57.8%	
Female (n = 66)	42.4%	57.6%	0.970
Pre-Existing Morbidity			
None (n = 109)	45.0%	55.1%	
Present (n = 206)	40.8%	59.2%	0.475
Registry Hospital Admission			
Direct from Scene (n = 178)	39.3%	60.7%	
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Hematologic[3]	**22**	**105**	**3**	**130**	**72**	**22.9%**
Cardiac	**19**	**71**	**20**	**110**	**75**	**23.8%**
GI/GU[4]	**12**	**49**	**10**	**71**	**52**	**16.5%**
Skin	**17**	**45**	**4**	**66**	**48**	**15.2%**
Psychiatric	**15**	**46**	**4**	**65**	**57**	**18.1%**
Total	**153**	**597**	**95**	**845**		
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[1] Column gives number of patients accounting for total number of complications by type

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		Coagulopathy	**4 (18.2%)**
		Deep Vein Thrombosis	**3 (13.6%)**
Cardiac	**19**	**Bradycardia/Dysrhythmia**	**8 (42.1%)**
		Shock (SBP < 80 mm Hg)	**2 (10.5%)**
		Cardiogenic Pulmonary Edema	**1 (5.3%)**
Psychiatric	**15**	**Depression**	**8 (53.3%)**
		Cognitive Deterioration	**2 (13.3%)**
Skin	**17**	**Sacral**	**6 (35.3%)**
		Heel/Scapular/operative wound	**4 (23.5%)**
GI/GU[3]	**12**	**Ileus/Hematuria**	**3 (25.0%)**
		Cholecystitis/Pancreatitis	**2 (16.7%)**

[1] Acute Lung Injury or Acute Respiratory Distress Syndrome

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		Wound Infection	**12 (9.2%)**
Hematologic	**105**	**Anemia[1]**	**48 (45.7%)**
		Coagulopathy	**16 (15.2%)**
		Thrombocytopenia	**13 (12.4%)**
		Deep Vein Thrombosis	**10 (9.5%)**
Cardiac	**71**	**Bradycardia/Dysrhythmia**	**29 (40.8%)**
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[1] Anemia is defined as a hematocrit ≤ 24% or hemoglobin concentration ≤ 8.0 g/dL

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Table 12. Leading causes of 95 severe complications by type

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		Pulmonary Embolus	**2 (5.0%)**
Cardiac	**20**	**Bradycardia**	**9 (45.0%)**
		Cardiac Arrest	**5 (25.0%)**
		Shock	**4 (20.0%)**
		Myocardial Infarction	**1 (5.0%)**
Infectious	**14**	**Pneumonia**	**9 (64.3%)**
		Wound Infection	**2 (14.3%)**
GI/GU	**10**	**Ileus**	**4 (40.0%)**
		Acute Renal Failure	**4 (40.0%)**
Skin	**4**	**Sacral**	**3 (75.0%)**
		Operative Wound	**1 (25.0%)**
Psychiatric	**4**	**Cognitive Deterioration**	**2 (60.0%)**
		Seizure	**2 (40.0%)**
Hematologic	**3**	**Anemia**	**1 (33.3%)**
		Thrombocytopenia	**1 (33.3%)**

NACTN: BUILDING A CLINICAL TRIALS NETWORK FOR SPINAL CORD INJURY

ROBERT G. GROSSMAN, ELIZABETH TOUPS, RALPH FRANKOWSKI, KEITH BURAU, SUSAN HOWLEY, for the NACTN Investigators*

The rapid advance of knowledge of the cellular and molecular responses of the spinal cord to injury has led to therapies that have improved functional recovery after spinal cord injury (SCI) in laboratory studies [1]. Some of these therapies have been used in small numbers of SCI patients [2]. However, most of these promising developments have not been brought to phase 2-3 clinical trials that have the design and statistical power to demonstrate efficacy because of the formidable organizational, regulatory and financial barriers that must be overcome to conduct such trials. In this chapter, we describe the steps in building the North American Clinical Trials Network (NACTN) for Treatment of Spinal Cord Injury. NACTN was created in 2004 with the support of the Christopher Reeve Foundation with the goal of overcoming these barriers. The Telemedicine and Advanced Technology Research Center (TATRC) United States Army Medical Research and Materiel Command (USAMRMC) of the Department of Defense has supported NACTN since 2006. Since 2007 Walter Reed Army Medical Center (WRAMC) has been part of NACTN with the goal of comparing the natural history of military and civilian injuries. NACTN's mission is to carry out clinical trials of the comparative effectiveness of new therapies for SCI using a consortium of neurosurgery departments at university-affiliated medical center hospitals with medical, nursing and rehabilitation personnel who are skilled in the evaluation and management of SCI. NACTN is the only standing network for clinical trials for SCI in North America. NACTN is composed of the following centers:

*NACTN Centers and Investigators

Clinical Centers

The Methodist Hospital, Houston – Coordinating Center
Principal Investigator, Robert G. Grossman, MD
Clinical Trials Manager, Elizabeth Toups, MS, RN, CCRP
The University of Texas-Memorial Hermann Hospital, Houston
Investigator, Michele Johnson, MD
The University of Virginia Hospital, Charlottesville
Investigators, Christopher I. Shaffrey, MD, John Jane, Sr. MD, PhD
The University of Toronto, Toronto
Investigators, Michael Fehlings, MD, PhD, Charles Tator, MD, PhD
The University of Louisville, Louisville
Investigators, Susan Harkema, PhD, Jonathan Hodes, MD
University of Maryland, Baltimore
Investigator, Bizhan Aarabi, MD
Walter Reed Army Medical Center
Investigator, Michael Rosner, MD
University of Miami, Miami
Investigator, James Guest, MD, PhD
Thomas Jefferson University, Philadelphia
Investigator, James Harrop, MD

Data Management Center
The University of Texas School of Public Health, Houston
Investigators, Ralph Frankowski, PhD, Keith Burau, PhD

Pharmacological Center
University of Houston, College of Pharmacy, Houston
Investigator, Diana Chow, PhD

BACKGROUND

An increasing number of clinical trial networks have been formed to study a wide variety of diseases [3]. This growth has been encouraged by the National Institutes of Health (NIH) [4] and the Institute of Medicine [5], and has been facilitated by internet communication, and the formation of societies and journals devoted to clinical trials [6]. The prototype of a clinical trial network for SCI was the National Acute Spinal Cord Injury Study (NASCIS) supported by the NIH, which published its first trial of corticosteroid therapy for SCI in 1984 [7]. NASCIS did not continue as a clinical trial organization for testing new therapies after the publication of its third corticosteroid

trial in 1997 [8]. In the past decade SCI clinical trial networks have been organized in Europe – the European Multicenter Study about Spinal Cord Injury (EMSCI) [9] and in Canada and the USA – the Surgical Treatment in Acute Spinal Cord Injury Study (STASCIS) [10] which was formed to study early surgical decompression of cord injuries. NACTN is collaborating with these organizations in studies of outcome after SCI.

STAGES IN THE DEVELOPMENT OF NACTN

NACTN has developed in 3 stages:

Stage 1: Recruitment of network components: Clinical Centers; Data Management Center; Pharmacological Center

Stage 2: Creation of a Data Registry of the natural history of SCI patients

Stage 3: NACTN's first clinical trial, of a neuroprotective drug, Riluzole

Each stage has involved multiple steps which are described below.

Stage 1: Recruitment of Network Components

Clinical Centers were sought that met the criteria of having:

1. Emergency transport systems providing rapid transport of SCI patients to a level 1 emergency department at the clinical center
2. An intensive care unit with neuro-intensivists
3. Neurosurgical, orthopedic, critical care and physiatry staff with research and clinical expertise in neuro-traumatology
4. A closely affiliated rehabilitation hospital
5. Mutual trust of the centers in each other based upon previous joint experience in research

The initial 5 clinical centers were expanded to nine centers between 2006 and 2008. Each center is led by a P.I. and at some centers a Co-P.I. Each clinical center has a study coordinator, usually a nurse-clinician and two clinical research assistants.

A Data Management and Statistical Center (DMC) was sought that had experience with large clinical trials, experimental design and familiarity with medical and governmental regulatory processes.

A Pharmacological Center was sought that had experience in pharmacokinetic and pharmacodynamic measurements of medications including, metabolism in animal and human studies.

Stage 2: Steps in the Creation of the Data Registry

Case report forms were developed to capture aspects of the medical history that have prognostic value. Forms were designed to obtain detailed information about complications that occur during the course of treatment. Clinical examinations were chosen to quantify the course of functional recovery in patients. A data archival system was developed to create a registry that can be interrogated to provide a historical control group to aid in clinical trial design.

Multiple meetings were held by the investigators to choose the data elements that would be collected and to design the case report forms. The investigators had the benefit of consulting existing SCI registries including those of the Model Spinal Cord Injury Systems [11], the International Spinal Cord Injury Data Set Elements that are currently being incorporated into the NIH – NINDS Common Data Elements[12] and those of STASCIS and EMSCI. Developing prognostic algorithms required collecting comprehensive data that included a medical history, medications, the radiology of the injury, medical and surgical treatment, the physiological

response to injury and complications, as well as neurological examinations from the earliest feasible time to examinations during rehabilitation and for a year after injury.

The case report forms are divided between measures obtained during the acute hospitalization following injury and functional outcome measures obtained during rehabilitation. The case report forms are available from the coordinating center to investigators who are developing clinical trials.

Acute Hospitalization Forms Data is collected under 12 headings:

Contact Sheet: Hospital Name, ID Number, Patient and Family Contact

Page 1 Demographic Data; Medical History

Page 2 Circumstances of Injury, Evacuation Details

Page 3 Initial Clinical Status; Glasgow Coma Score (GCS); Abbreviated Injury Score (AIS)

Page 4 A,B ASIA (American Spinal Injury Association) Motor, Sensory Scales at <72h and 2 weeks

Page 5 Type of Neurological Injury; Type of Bony Injury

Page 6 Imaging Cord and Canal Diameters: CT; MRI; CT/Myelogram

Page 7A,B Non-Operative Treatment: Medical; Traction - Reduction

Page 8A,B Posterior Surgical Treatment: Procedural Details; Levels

Page 9A,B Anterior Surgical Treatment: Procedural Details; Levels

Page 10A-D Complications: Cardiac; Pulmonary; Hematological; GI/GU; Infection; Skin; Psychiatric

Page 11 Acute Hospitalization Outcome Summary

Rehabilitation and Long-Term Follow-Up Forms Functional outcomes are measured at specified times:

ASIA Scale (American Spinal Injury Association) at 3, 6 and 12 months after injury

FIM (Functional Independence Measure) at Acute Discharge, Rehabilitation Discharge, 3, 6, 12 months

SCIM (Spinal Cord Independence Measure) at Acute Discharge, Rehabilitation Discharge, 3, 6, 12 months

WISCI-II (Walking Index for Spinal Cord Injury) at Acute Discharge, Rehabilitation Discharge, 3, 6, 12 months

Data Quality Control: The NACTN Manual of Operations (MOO)

The MOO was written to serve as a guide for completing the data forms. It is a 55 page document that summarizes the research protocol and inclusion/exclusion criteria, and gives explicit instructions about each form. The center study coordinators are also in frequent telephone and e-mail communication with the Clinical Coordinating Center and the Data Management Center to clarify questions that arise.

Data Form Design and Transmission of Data from the Centers to the DMC

A multi-part paper/optical scanning system was judged to be the best initial system for data collection since it would allow for multiple protocol iterations and adjustments required as experience was gained in data collection. The data collection system is a high security system. Data from patients that is submitted to the DMC is identified only by a code number for the center, and a number for the patient. Each data collection form was designed in Verity TeleForm software that uses an optical recognition system to digitally capture paper-based images and their content by scanner and allows customized field-by-field data verification. The data processing

activities are managed by an integrated data system that uses TeleForm for data entry and verification and then uploads data to a secure Structured Query Language (SQL) server which simultaneously generates Microsoft (MS) Access files and tables. This integrated system is augmented by a parallel data quality control system.

The NACTN registry receives completed paper forms via mail from the clinical centers. Data forms are reviewed for completeness before being sent to the optical scanner. Forms are held by the electronic data processing system until they are audited by a verification program that allows field-by-field data verification. Each scanned form is maintained in the system as a TIF file allowing for computerized images of the data forms to be stored electronically. Once the data forms have been verified, the data go to a secure password-protected SQL server database. This server is backed up nightly with archival storage maintained at two alternate off-site locations. Once data have been committed to the data tables, the data are subjected to logic and edit checks as well as cross-checks between different form types. A separate file tracks each batch of data entered through the scanning system. Additionally, a correction database has been developed to document data change and/or data transactions that result from logic/error checks or updates received from clinical centers. Computer programs have also been written to generate one-page patient narrative reports. Export procedures were developed to convert data to SAS and State formats to facilitate statistical analyses. Programs have been developed to tabulate summary statistics and generate reports. All steps from data logging, entry, committing to SQL and MS Access, editing, reporting and summary analysis have been closely documented and are known by multiple members at the DMC. This redundancy and shared responsibility reduces the turn-around time from receipt of forms to generation of data editing reports sent back to the

clinics. As patients are enrolled in the registry, the quality of data on key measures is evaluated and, when necessary, measures are refined or clarified in the DMC Manual of Operations.

Steps in the Initiation of the Data Registry

Protocol Development A protocol was written that stated the goals of the registry, the inclusion/exclusion enrollment criteria, the process of data collection and the precautions to ensure patient protection and the security of the data.

An Informed Consent Form (ICF) and an Explanatory Brochure for patients were written.

IRB Approval of the Protocol and the ICF was obtained from the Institutional Review Boards of the centers, and from the Human Research Protection Office (HRPO), Department of Defense.

Training in Data Collection Two workshops were held for the investigators, nurse clinicians, physiatrists and coordinators at which they received training in the ASIA examination and in the data collection methodologies.

Data in the Registry The registry has enrolled 414 patients with ASIA grades A to E, from October, 2005 to October, 2010.

Stage 3: NACTN's Initial Clinical Trial: "A Phase 1 Study of the Safety and Pharmacokinetics of Riluzole in Patients with Traumatic Acute Spinal Cord Injury"

Riluzole is a neuroprotective drug that blocks glutamate-mediated sodium and calcium ion entry into neurons and glia and blocks glutamate release from presynaptic terminals. It has been shown to be effective in limiting traumatic damage to the spinal cord in laboratory studies[13]. Riluzole presented several advantages as a therapy in NACTN's first trial in which the working of the network was being tested. Riluzole is used clinically in the treatment of amyotrophic lateral sclerosis (ALS) [14]. It is relatively inexpensive; it can be given orally; it has a

favorable safety profile in ALS patients; it can be measured in blood and CSF and costs are reasonable for laboratory studies that are necessary for monitoring its use.

Research Design and Methods

The initial phase of the study is a multi-site, single arm active treatment pilot study involving 36 subjects. If the rate of adverse effects in the initial trial group is no greater than that in the NACTN data base, a phase 2 study of a larger number of patients will be undertaken as a comparative efficacy trial. Features of the Riluzole protocol are the detail in which possible adverse effects of the therapy are being studied, and the use of pharmacokinetic and pharmacodynamic data and its correlation with adverse effects and efficacy. Previous studies of drug therapy for SCI have not measured blood and cerebrospinal fluid levels of the therapeutic drugs to see if effective or toxic levels of drug were reaching the spinal cord and brain.

Multiple steps were involved in planning and in implementing the Riluzole trial which enrolled its first patient on April 12, 2010.

Selection of a Therapy and Development of the Protocol

1. Selection of Riluzole after discussion by the investigators of candidate therapies.
2. Writing the research protocol
3. Creation of a schedule of events (SOE), a day-by-day hourly schedule of tests, procedures, laboratory work, rules for drug administration
4. Modifying elements of the registry case report forms for the requirements of the trial
5. Designing case report forms (CRF)
6. Writing a Manual of Operations and patient brochure
7. Writing the Informed Consent Form
8. Setting up the study database at the DMC

9. Developing new methods of measuring Riluzole in plasma and CSF

Trial Initiation: Compliance with regulatory requirements:

1. Approval of the research protocol by the HRPO, DOD
2. Harmonization of the IRB requirements of each center with requirements of the HRPO; final approval of harmonized protocol and ICF by each IRB
3. Appointment of a central trial monitor, a distinguished physiatrist at a university not affiliated with any of the centers. For a Phase 2 study a data monitoring and safety committee will be established
4. Appointment of a local trial monitor at each clinical center
5. Trial initiation meeting of all investigators and coordinators. Intensive two day review of the protocol, the schedule of events, the rules and procedures for reporting adverse events, interim analysis and stopping rules
6. Writing and signing of site agreements between the trial sponsor and the network centers The site agreements are a contract setting forth the statement of work, the responsibilities of the center and deadlines for completion of work

Trial Conduct at Each Center

1. Creation of patient data binders
2. Creation of regulatory binders
3. At the Coordinating Center, files of all regulatory documents and files of adverse events

Appointment of a Site Monitor

An individual trained in monitoring clinical trials who has been conducting on-site monitoring at each clinical center and reviewing CRFs, regulatory documents, source

documentation, adherence to protocol and drug accountability in accordance with federal regulations and Good Clinical Practices (GCP)

Insuring Communication between Network Centers:

1. Writing a Governance Manual
2. Forming committees: Executive; Selection of Therapies; Publications; Data Management
3. Forming Neurological Outcomes Assessment Task Force, led by Dr. Susan Harkema, to develop improved quantitative outcome measures
4. Conducting monthly conference calls by the investigators and coordinators, the committees and the NOA Task Force
5. Posting of NACTN documents and communications on a FTP website

CONCLUSIONS:

Pearls (1) Clinical trial organization and implementation requires a fusion of many disparate elements in our complex, non-integrated medical system.

(2) The most important elements in building a clinical trials network is the dedication of the investigators and the clinical and scientific staff and the willing participation of patients, to whom we owe our gratitude.

(3) Current thinking is that a trial of a new therapy for SCI will require a cohort of 200-250 patients to achieve statistical significance. Depending on the therapy the cost of such a study can be in the millions of dollars. An urgent question is how to apportion these costs between governmental and private funding agencies, pharmaceutical companies, voluntary health organizations, not-for-profit hospitals, philanthropy and health insurers. NACTN has combined support from multiple sources to develop a clinical trial network. We believe that a parallel

effort of development of consortia of funding sources is required to support clinical trials of new therapy to improve the outcome of SCI.

Pitfalls (1) Randomized multicenter clinical trials require extensive planning that frequently takes 1-2 years to complete. Additional time is required for harmonization of the protocol with specific requirements that individual IRBs request, and which must be acceptable to all of the other IRBs, requiring frequent protocol revisions and IRB filings.

(2) The complexity of planning and initiating multi-center clinical trials and the cost of such trials have been major factors in preventing new therapies from being brought from the laboratory into clinical practice.

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Additional support provided by Mission Connect, a project of TIRR Foundation

“A Phase 1 Clinical Trial of Riluzole, a Neuroprotective Drug, for Acute SCI”

North American Clinical Trials Network for Treatment of Spinal Cord Injury (NACTN)

Robert G. Grossman, MD, Elizabeth G. Toups, MS, RN, CCRP, for the NACTN Investigators

Riluzole is a neuroprotective drug that blocks glutamate-mediated sodium and calcium ion entry into neurons and glia and blocks glutamate release from presynaptic terminals. It has been shown to be effective in limiting traumatic damage to the spinal cord in laboratory studies (Schwartz and Fehlings, 2001). Riluzole presents several advantages as a therapy for acute SCI. Riluzole is already used clinically, in the treatment of amyotrophic lateral sclerosis (ALS). It is relatively inexpensive; it can be given orally; it has a favorable safety profile in ALS patients; it can be measured in blood and CSF and costs are reasonable for laboratory studies that are necessary for monitoring its use.

The primary goals of the study are to:

1. Evaluate the safety of the drug
2. Obtain information about the pharmacodynamics and pharmacokinetics of its administration in acute SCI
3. Relate pharmacological data to complication rates
4. Collect information about efficacy as measured by American Spinal Injury Association ASIA Score, ASIA Impairment Grade and Brief Pain Inventory

RESEARCH DESIGN AND METHODS

The initial phase of the study is a multi-site, single arm active treatment pilot study involving 36 subjects. If the rate of adverse effects in the initial trial group is no greater than that in the NACTN database, a phase 2 study of a larger number of patients will be undertaken as an efficacy trial. Features of the Riluzole protocol are the detail in which possible adverse effects of the therapy are being studied, and the use of pharmacokinetic and pharmacodynamic data and its correlation with adverse effects and efficacy. Previous studies of drug therapy for SCI have not measured blood and cerebrospinal fluid levels of the therapeutic drugs to see if effective or toxic levels of drug were reaching the spinal cord and brain.

The Riluzole trial enrolled its first patient on April 12, 2010 at the University of Maryland Shock Trauma Hospital.

NACTN Clinical Centers / Data Management Center / Pharmacological Center:

1. **The Methodist Hospital, Houston**
   Principal Investigator, Robert G. Grossman, M.D.
2. **University of Texas-Memorial Hermann Hospital, Houston**
   Investigator, Michele Johnson, M.D.
3. **University of Virginia Hospital, Charlottesville**
   Investigator, Christopher I. Shaffrey, M.D.
4. **University of Toronto**
   Investigator, Michael Fehlings, M.D., Ph.D.
5. **University of Louisville, Louisville**
   Investigator, Susan Harkema, Ph.D.
6. **University of Maryland**
   Investigator, Bizhan Aarabi, M.D.
7. **Walter Reed Army Medical Center**
   Investigator, Michael Rosner, M. D.
8. **University of Miami, Miami**
   Investigators, James Guest, M.D., Ph.D.
9. **Thomas Jefferson University, Philadelphia**
   Investigator, James Harrop, M.D.

Data Management Center
University of Texas School of Public Health, Houston
Ralph Frankowski, Ph.D.

Pharmacological Center
University of Houston, College of Pharmacy, Houston
Diana Chow, PhD

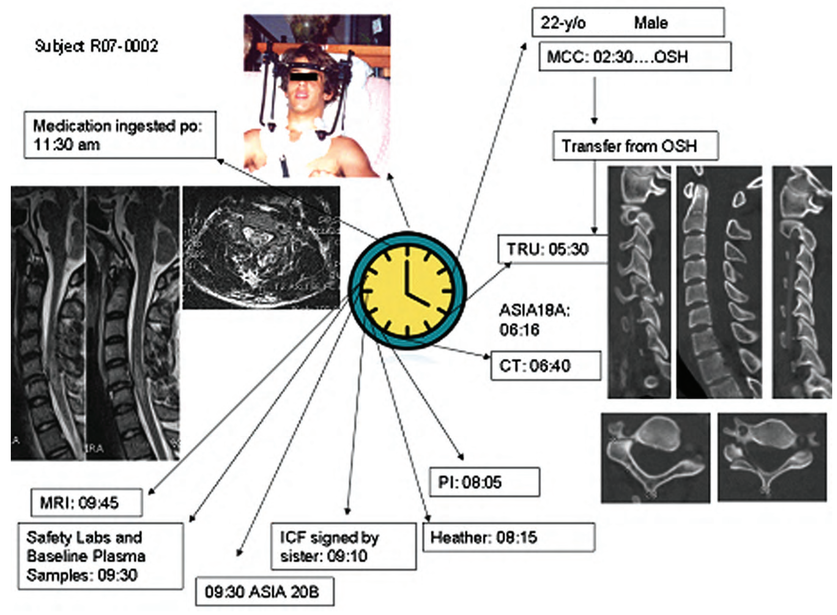
PRELIMINARY ANALYSIS

At the present time, 22 patients are enrolled. The University of Maryland and Thomas Jefferson University have each enrolled 8 patients. The University of Virginia has enrolled 4. The University of Texas – Memorial Hermann Hospital and the University of Louisville have each enrolled 1 patient. Pharmacological analysis on 16 patients indicates expected blood concentration of Riluzole. There is no evidence of toxicity at this point in time.

Disclosures: N/A

Support: NACTN is supported by the Christopher Reeve Foundation, the Department of Defense - U.S. Army Medical Research and Materiel Command and Mission Connect of the TIRR Foundation.

TIMELINE OF INITIATION OF TREATMENT OF PATIENT #2



NACTN

North American Clinical Trials Network for Treatment of Spinal Cord Injury

Supported by
Reeve Foundation
Department of Defense
Mission Connect of the TIRR Foundation

REEVE FOUNDATION
NORTH AMERICAN
CLINICAL TRIALS NETWORK®

CLINICAL TRIALS IN SPINAL CORD INJURY
THE ROAD AHEAD

Today there is hope for improved functional recovery after SCI. This hope is based upon new understanding of the cellular and molecular processes of injury and repair of the spinal cord.

ClinicalTrials.gov
SCI Studies

- 318 Total
- 103 Active
- 3 Phase I Multicenter Trials with the prospect of going into Phase II Randomized Controlled Trials

  - NACTN - Riluzole
  - Novartis - Anti-Nogo ATI355
  - Geron – GRNOPC 1

CAUTION!

All randomized prospective clinical trials of therapy for SCI that had improved outcome in animal models of SCI have failed to demonstrate efficacy in man.

OBSTACLES TO PROGRESS IN CLINICAL TRIALS

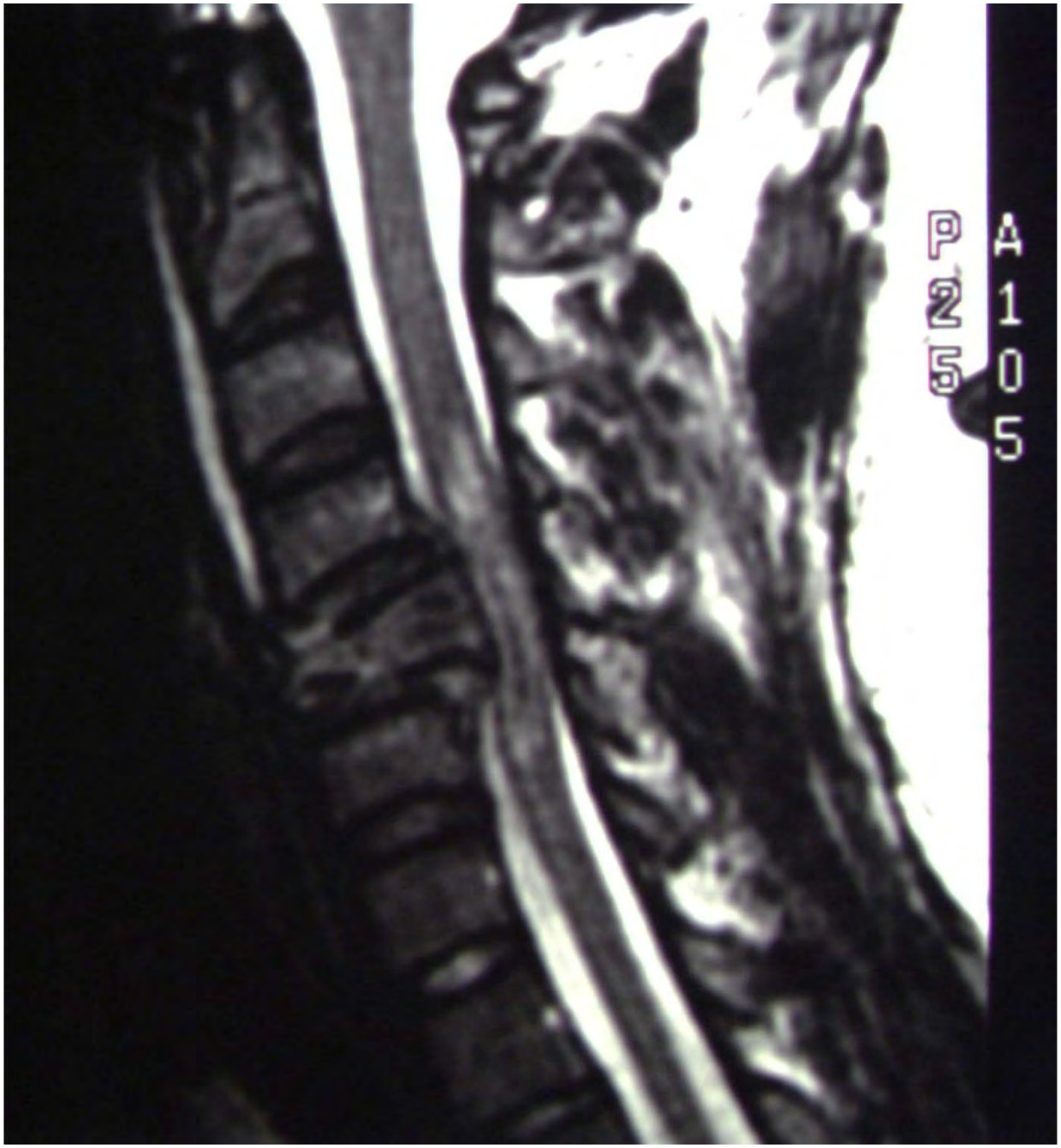
BIOLOGICAL

The heterogeneity of the biological processes in SCI and their evolution over time

SOCIAL

The formidable organizational, regulatory and financial requirements for carrying out SCI clinical trials

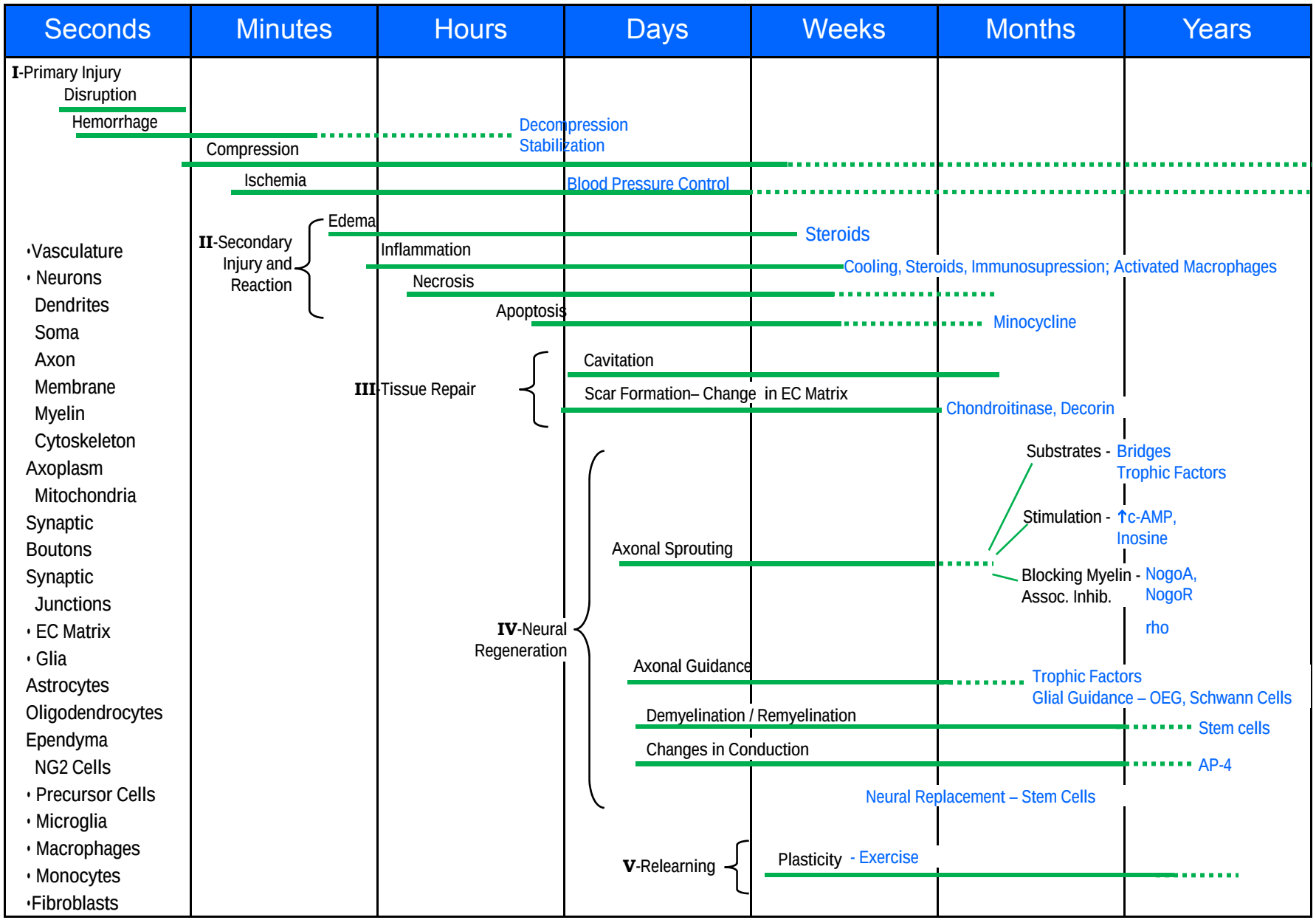
BIOLOGY OF CORD INJURY



THE BIOLOGY OF CORD INJURY

A process that develops over days, months and years, involving traumatic and ischemic cell damage and death, invasion by inflammatory cells, clearing of debris, abortive repair of the structural scaffolding and blood supply of the spinal cord, neural sprouting and forming of new circuits under the influence of neural activity

Mechanisms of SCI and Their Temporal Dimensions



CLINICAL VARIABLES THAT DETERMINE OUTCOME

1. Age of the patient
2. Pre-existing medical conditions
3. Genetic polymorphisms
4. Associated injuries, hypoxia, hypotension
5. Variations in the pathology and extent of injury – hematoma, compression, ischemia
6. Variability of the medical and surgical therapy
7. Time to treatment
8. Medical complications that develop during treatment, particularly infections

Requirements for a Clinical Trial in SCI

1. **Proof of Principle in Laboratory Studies; Replication of efficacy in different species**
2. **Design of Human Studies**

   A. Recognizing biological and clinical variability to enable stratification of patients

   B. Obtaining accurate and reproducible measurement of small changes in clinical outcomes, particularly of the course of motor recovery

   C. Developing outcome measures that are sensitive to the biological effects of the therapy. For example, for a cervical injury treated with a regenerative therapy improvement in hand function is more likely to occur than in leg function, and sensitive tests of hand function will be required to detect improvement

3. **Statistical Analysis of Variability of Outcomes - Calculating the number of patients required to detect a particular outcome**

4. **Phase I – Safety study: pharmacokinetics and toxicology. Limited number of patients, often 36 – 48 subjects. Can be single or multi- center**

… The Major Step—The Bottleneck--

5. **Phase II/III Efficacy Study – Current thinking is of 150 – 250 subjects for SCI trial. Multicenter to obtain sufficient numbers of SCI**

BUILDING A CLINICAL TRIALS NETWORK: NACTN

With these requirements in mind, in 2004, neurosurgeons from five medical centers created a clinical and research infrastructure to conduct trials of new therapies for SCI. The consortium chose as its name the North American Clinical Trial Network (NACTN)

NACTN Centers and PIs

1. **The Methodist Hospital, Houston**

   Robert G. Grossman, M.D.

   Project Manager, Elizabeth Toups, M.S., R.N., CCRP

2. **University of Toronto, Toronto**

   Michael Fehlings, M.D., Ph.D.,Charles Tator, M.D., Ph.D.

3. **University of Texas, Houston**

   Michele Johnson, M.D.

4. **University of Virginia Hospital, Charlottesville**

   Christopher I. Shaffrey, M.D.

5. **University of Louisville, Louisville**

   Susan Harkema, Ph.D., Jonathan Hodes, M.D.

6. University of Miami, Miami

James D. Guest, M.D., Ph.D.

7. Thomas Jefferson University, Philadelphia

James Harrop, M.D.

8. University of Maryland, Baltimore

Bizhan Aarabi, M.D

9. Walter Reed Army Medical Center, Washington DC

Michael Rosner, M.D.

Pharmacological Center

University of Houston, College of Pharmacy

Diana Chow, Ph.D

Management and Statistical Coordination:

University of Texas School of Public Health – Data Management and Statistical Coordinating Center

Ralph Frankowski, Ph.D.

NACTN Centers

NACTN INTERACTIONS-

The basic science laboratories of the Reeve Foundation International Research Consortium

European Multicenter Study on Spinal Cord Injury (EM-SCI)

Surgical Trial in Acute Spinal Cord Injury Study (STASCIS)

Spinal Cord Outcomes Partnership Endeavor (SCOPE)

International Campaign for Cures of Spinal Cord Injury Paralysis (ICCP)

NIH NETT (National Institutes of Health Neurological Emergency Treatment Trials Network)

NINDS

Steps in Development of NACTN

Stage I

- Expansion to 9 clinical centers
- Developing the Pharmacological Center
- Developing the Data Management Center
  - Writing the Data Registry Protocol and Manual of Operations (MOO)
    - Selecting data elements & outcome measures
    - Creating case report forms (CRF)
    - Creating data transmission system between the clinical centers and DMC
- Training of personnel on the protocol, use of the CRFs and performing the ASIA and other examinations
- Developing a data base of the natural history of SCI
- Determining rates of complications during acute hospitalization

Steps in Development

Stage II

Creating the Neurological Outcomes Assessment Task Force (NOA) to develop sensitive and accurate outcome measures

Stage III

Developing and starting NACTN's first clinical trial: A Phase I Study of the Safety and Pharmacokinetics of Riluzole in Patients with Traumatic Acute Spinal Cord Injury

Stage IV

In 2011, adding additional civilian and military hospitals

NACTN TASKS - FROM CONCEPT TO CLINICAL TRIAL

1. **Obtain Data on the Natural History of SCI**
   - **A. NACTN Registry Data**
   - **B. Distribution of Outcomes With Current Standard of Care Treatment**
   - **C. Quantitative Measurements of Motor Outcome**
   - **D. Rates of Complications -Significance for trials**
2. **NACTN's first trial: RILUZOLE - a NeuroprotectiveTherapy**

1A. NACTN DATA ON THE NATURAL HISTORY OF SPINAL CORD INJURY

As of October 2010, 742 patients with acute spinal cord injuries have been screened at the time of admission to NACTN centers and 392 patients have been enrolled into the NACTN database.

The database contains sequential neurological examinations, the radiological characteristics of the injury to the spinal cord and to the vertebral column and detailed information about complications, with a follow-up period of a year after injury.

North American Clinical Trials Network

Patient Demographics

Characteristic	Number (N=392)	Percent
Gender		
Male	311	79
Female	81	21
Age[1] (yrs)		
< 20	29	7
20-65	314	80
>65	49	12
Race		
White	299	76
Other	93	24

[1]Median age at injury = 44.0 yrs of age

Injury Region[1]	Number	Percent
Cervical	295	75
Thoracic	70	18
Lumbar/Sacral	24	6
SCIWORA	3	1

ASIA Grade at Admission

ASIA Score	Number	Percent Total
A	132	34
B	53	14
C	49	15
D/E	133	34
Unknown	25	6
TOTAL	392	100

Surgeries by ASIA Grade

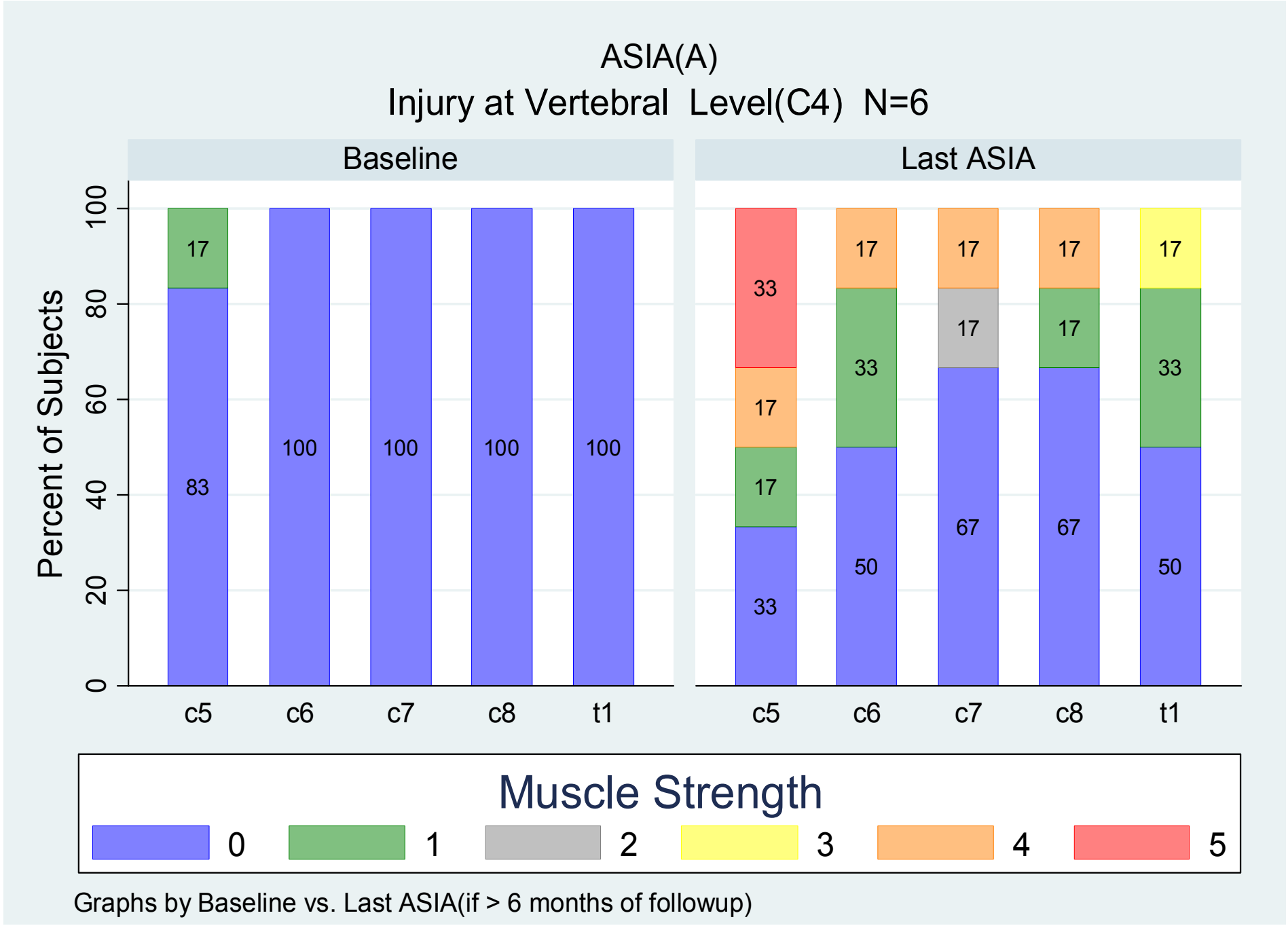
	Surgeries (N)					
ASIA Admit	Posterior	Anterior	Both	None	TOTAL	% Operated
A	60	16	21	10	107	91
B	18	9	7	6	40	85
C	20	7	2	6	35	76
D/E	32	36	12	28	108	74
Unknown	8	8	3	8	27	70
TOTAL	138	76	45	58	317	82

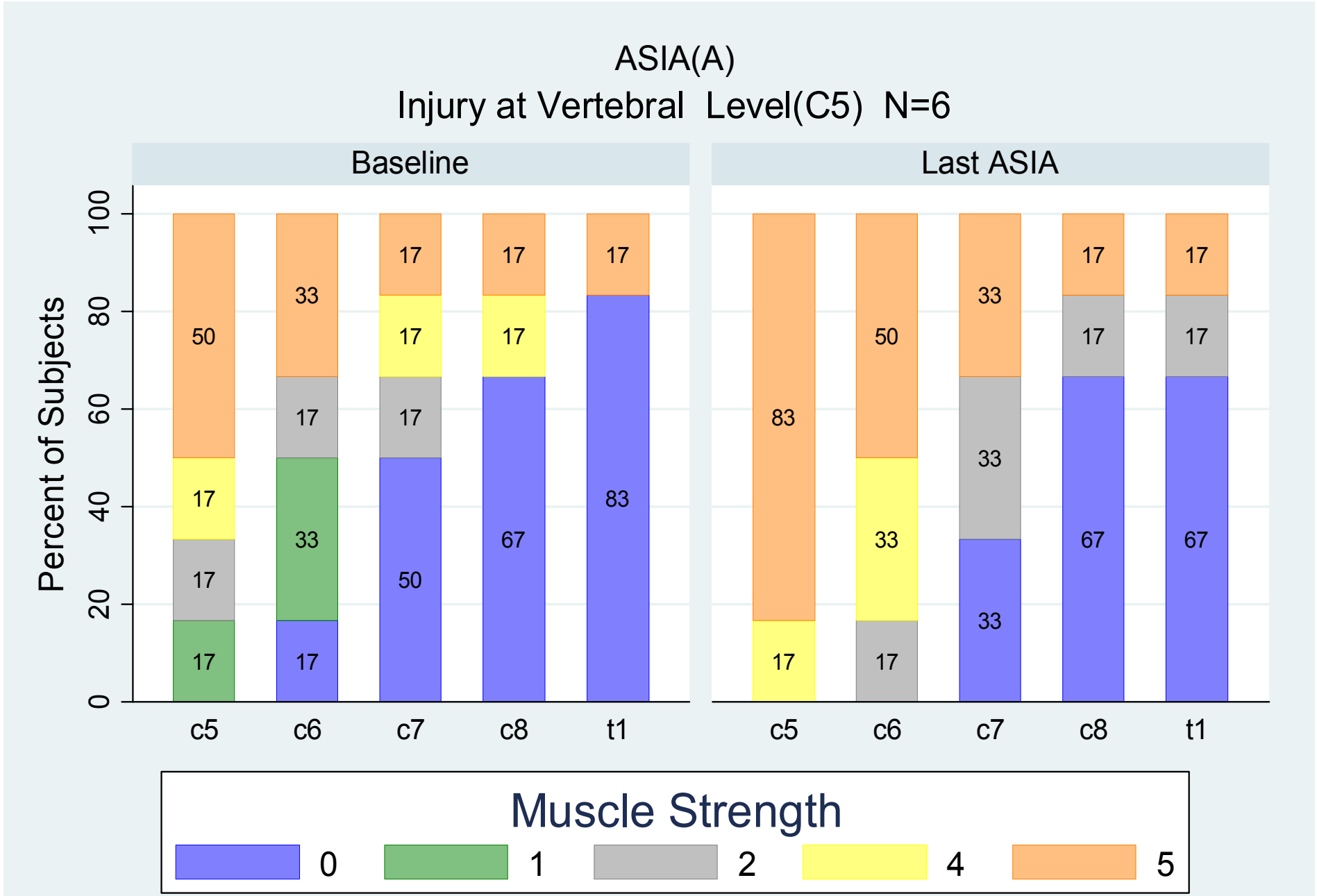
1B. METHODS OF DISPLAYING AND ANALYZING THE DISTRIBUTION OF OUTCOMES

1B. Distribution of Outcomes With Current Standard of Care

Conversion of ASIA Grade during Acute Hospitalization: ASIA Impairment Scale on Admission and at Discharge

	ASIA Discharge					
ASIA Admit	A	B	C	D/E	Unk	
A -132	106	9	4	0	13	
B -53	6	30	13	2	2	
C -49	1	1	30	16	1	
D/E-133	0	0	4	127	2	
Unknown-25	5	2	3	5	10	
All -392	118	42	54	150	28	

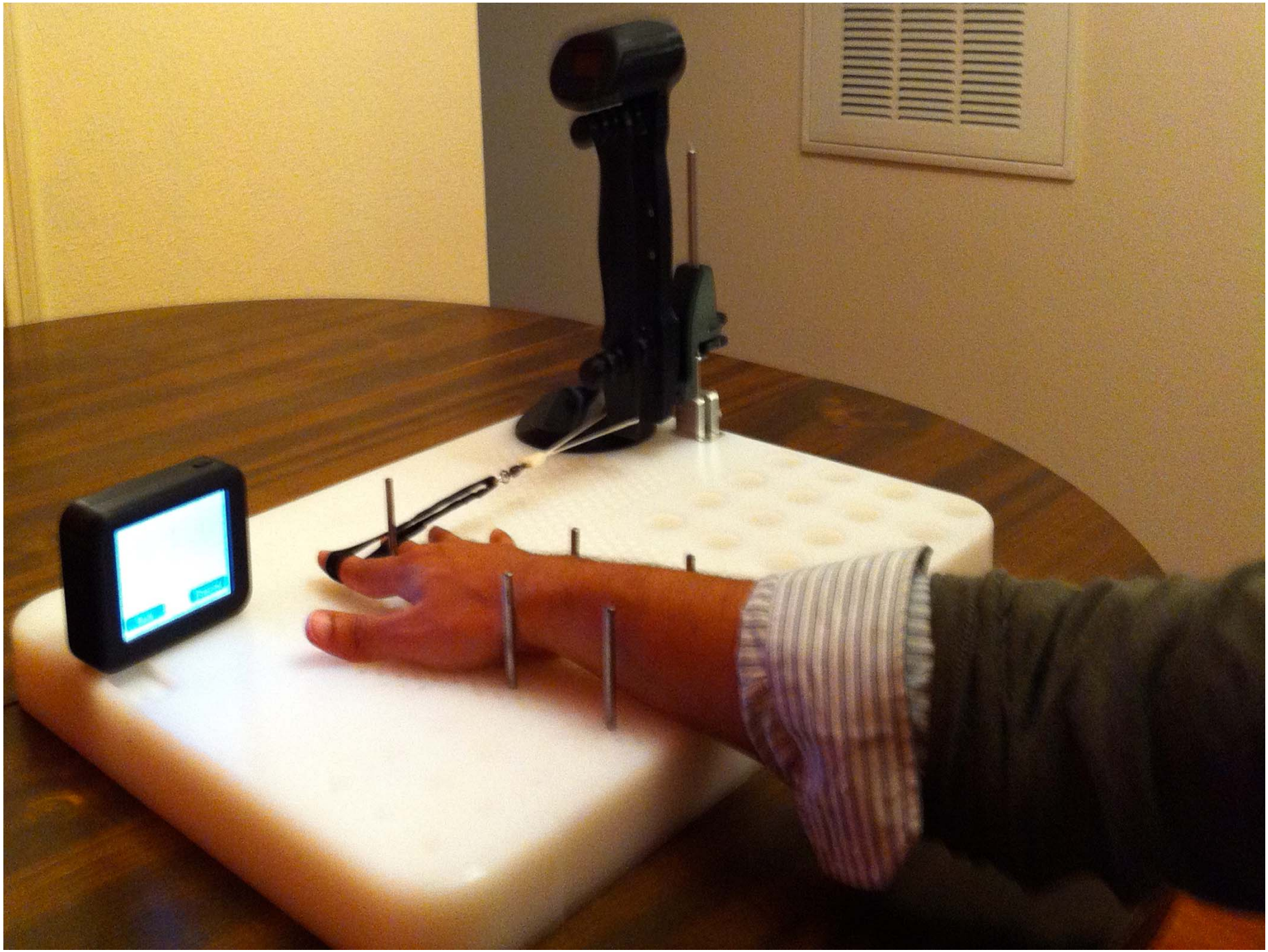

ASIA(A)
Injury at Vertebral Level(C4) N=6
Baseline
Last ASIA
Percent of Subjects
0
20
40
60
80
100
17
83
100
100
100
100
33
17
17
33
17
17
17
33
17
17
17
33
67
67
50
50
c5
c6
c7
c8
t1
c5
c6
c7
c8
t1
Muscle Strength
0
1
2
3
4
5
Graphs by Baseline vs. Last ASIA(if > 6 months of followup)

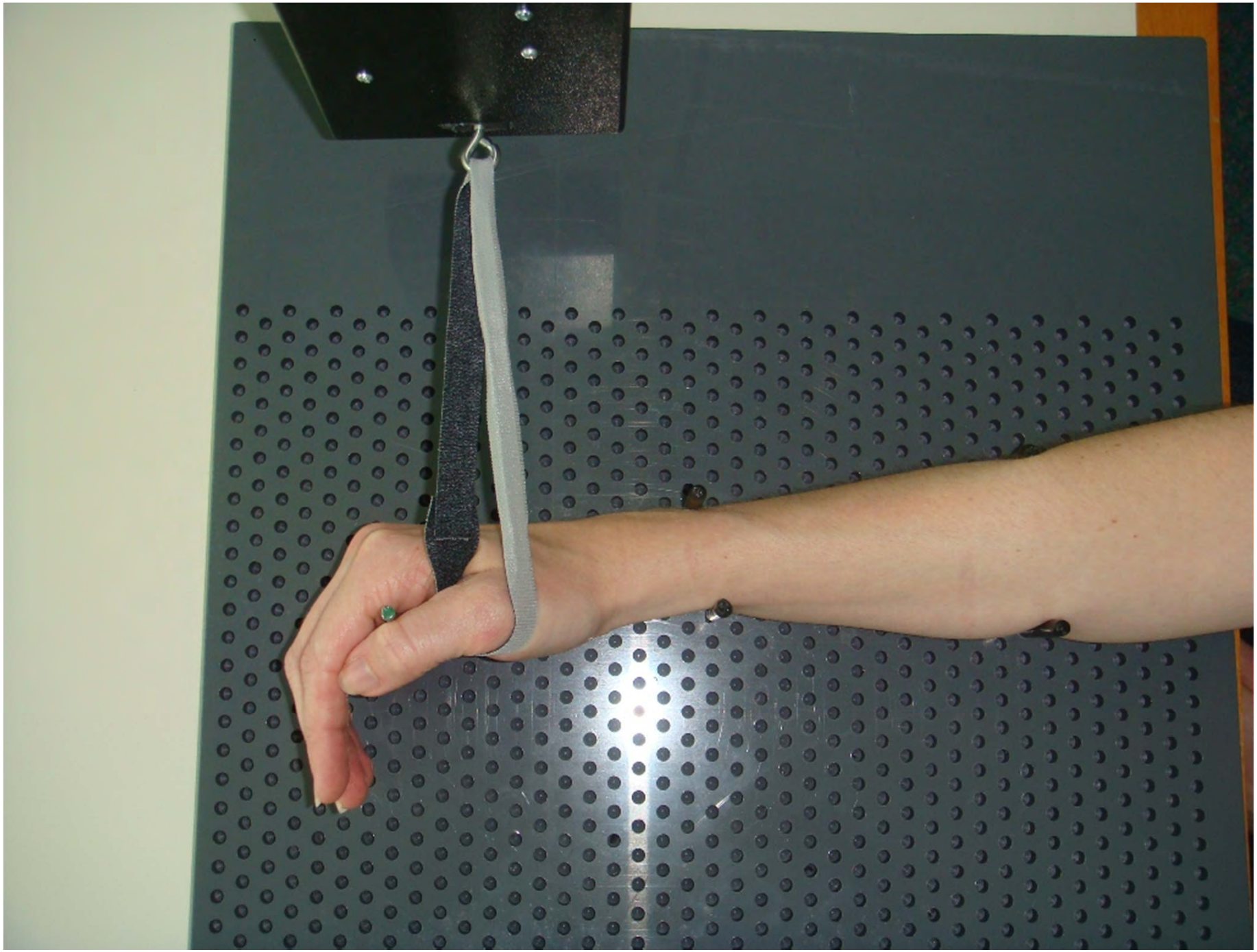


Graphs by Baseline vs. Last ASIA(if > 6 months of followup)

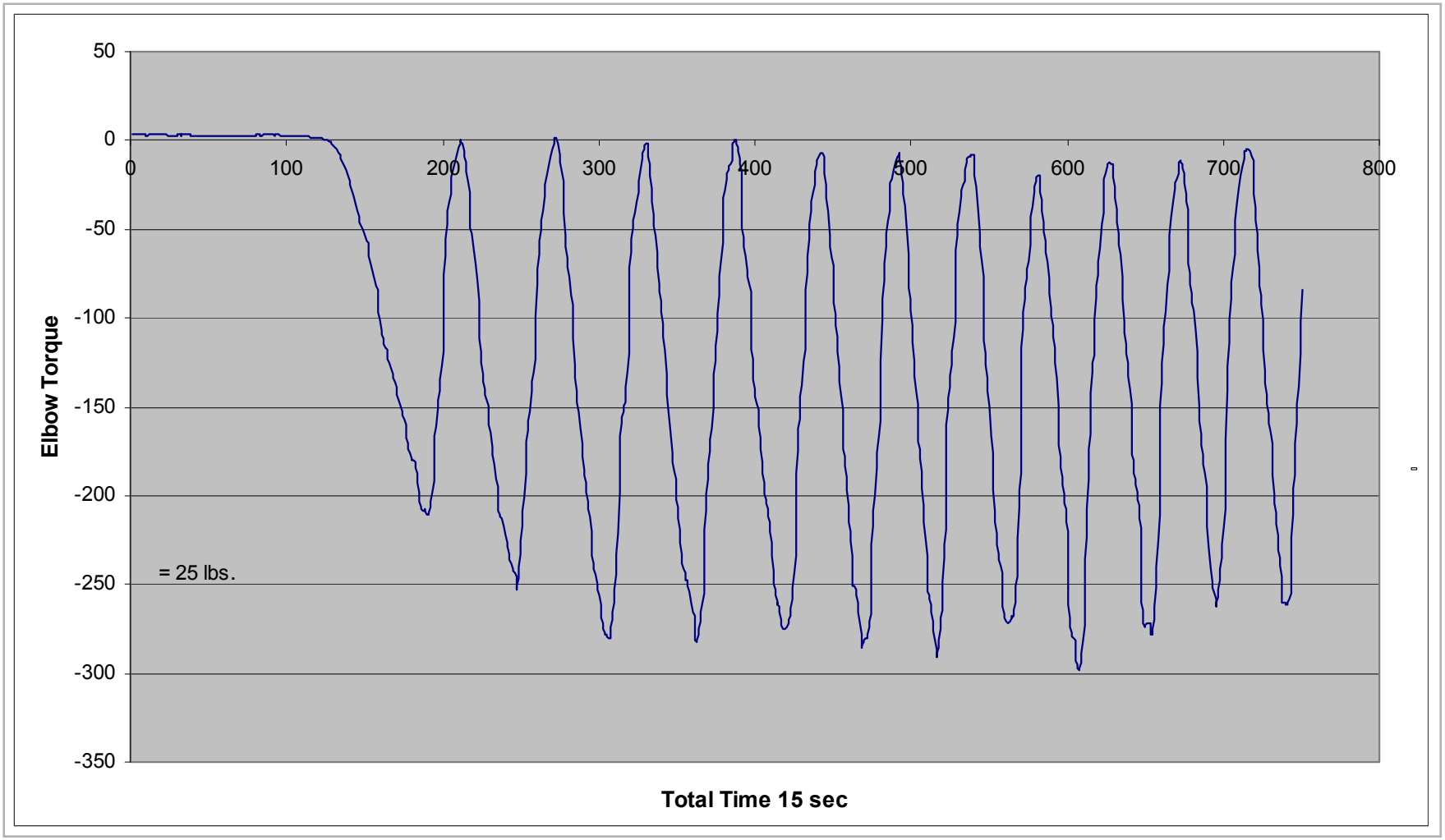
1C. Quantitative Measurement of Motor Outcome: Peg Restrained Intrinsic Muscle Evaluator (PRIME)

Gloria Gogola, M.D., Shriners Hospitals for Children





Triceps Max Torque



1D. RATE OF ACUTE COMPLICATIONS AFTER SCI

Establishment of rates of medical and surgical complications in SCI patients receiving the best standard therapy is crucial for validating Phase I safety trials of new therapies.

Phase I studies typically do not have a control group and do not involve a large number of patients. Statements are often made in safety trials that few, or no adverse effects were seen that were due to the therapy.Such statements cannot be made without reference to a historical control group such as has been compiled by the NACTN database.

This data is available for use by all SCI clinical trials.

Frequency of Complications within Categories

306* SCI Patients with 736 Complications (excluding 11 deaths)

Complications (N)	Common	Moderate frequency	Infrequent
Pulmonary (185)	Ventilatory failure 32.4% (60)	Pleural effusion 24.9% (46)	Other pulmonary 11.9% (22)
Infection (155)	Pneumonia 41.3% (64)	Urinary infection 25.2% (39)	Empyema 16.8% (26)
Hematological (123)	Anemia 49.6% (61)	Other hematology 19.5% (24)	Coagulopathy 13.8% (17)
Cardiac (85)	Bradycardia 31.8% (27)	Shock 25.9% (22)	Other cardiac 16.5% (14)
Psychiatric (64)	Depression 65.6% (42)	Other psychiatric 20.3% (13)	Cognitive deterioration 0.9% (6)
Skin (63)	Other skin 41.3% (26)	Sacral 39.7% (25)	Heel 9.5% (6)
GI/GU (61)	Other GI/GU 67.2% (41)	GI hemorrhage 9.8% (6)	Hematuria 8.2% (5)

* Number of patients includes 141 with no complications

The findings are important for designing trials for SCI. Therapies that impair the immune response and lead to infections and therapies that cause pulmonary, cardiac and hematological disturbances may not be feasible orwill require intensive early monitoring to prevent or reduce adverse effects.

The data serve as a benchmark for evaluating the safety of new therapies in SCI.

2. RILUZOLE AS A NEUROPROTECTIVE AGENT NACTN'S FIRST CLINICAL TRIAL

- **Blocks glutamate-mediated sodium and calcium ion entry into neurons and glia**
- **Shown to be effective in limiting traumatic damage to the spinal cord in laboratory studies (Schwartz and Fehlings, 2001)**
- **Currently used in clinical practice in treatment of amyotrophic lateral sclerosis (ALS)**

Aims of the Phase I Riluzole Study

To evaluate the drug's safety

Obtain information about the pharmacodynamics and pharmacokinetics of its administration in acutely injured SCI patients

Relate pharmacological data to adverse events

Riluzole Trial- Steps

Writing Riluzole Protocol, Investigational Guide and Procedural Guide

Development of CRFs

Analytic methods to measure Riluzole in plasma and spinal fluid

Recruitment of central medical monitor, Steve Williams, MD, Boston Univ

Institutional Review Board (IRB) approval at each clinical center and from the DOD Office of Research Protection

NACTN-wide personnel training in the Riluzole protocol and clinical examinations

Site Initiation/Training meeting Houston, January 14-15, 2010

Enrolling the first patient in the trial on April 12, 2010 at theShock-Trauma Hospital, University of Maryland, Baltimore - Dr. Bizhan Aarabi

Subject R07-0002

22-y/o Asian Male

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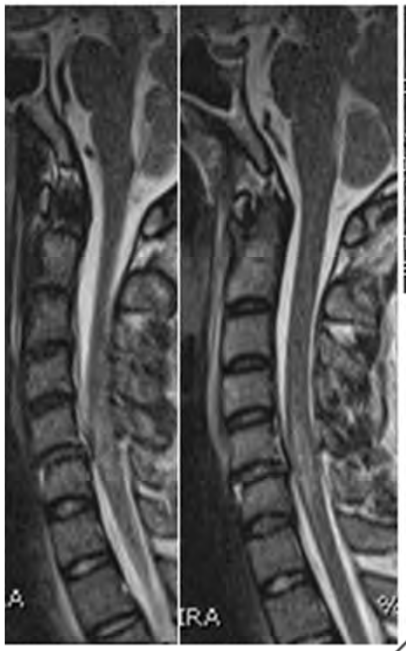
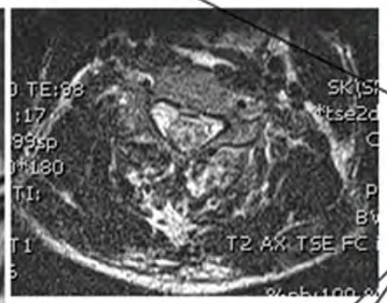
Transfer from OSH

Medication ingested po: 11:30 am





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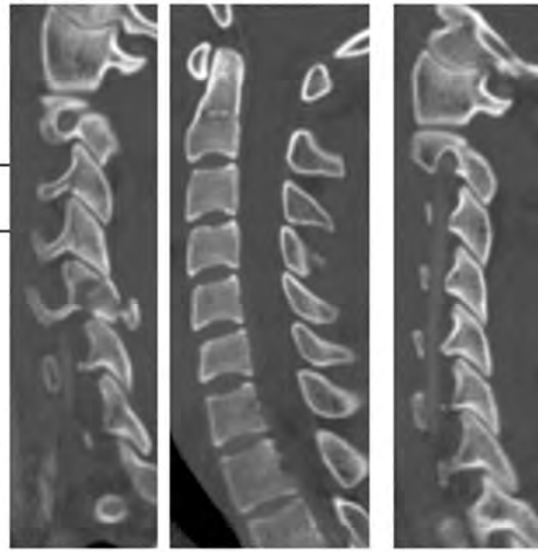

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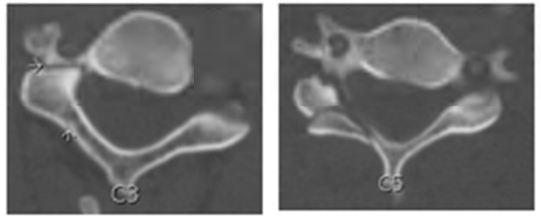

PI: 08:05

MRI: 09:45

Safety Labs and Baseline Plasma Samples: 09:30

ICF signed by sister: 09:10

Heather: 08:15

09:30 ASIA 20B

CURRENT STATUS OF THE RILUZOLE TRIAL

At the present time 23 patients are enrolled.

Pharmacological analysis has been completed on 16 patients. Expected blood concentration of Riluzole have been acheived. There is no evidence of toxicity at this point.

LESSONS FROM THE ROAD TRAVELLED
and for
THE ROAD AHEAD

Current thinking is that a trial of a new therapy for SCI will require 150 – 250 patients to achieve statistical significance. Depending on the therapy the true cost of such a study can be in the millions of dollars. An urgent question is how to apportion these costs between government and private funding agencies, pharmaceutical companies, voluntary health organizations, not-for-profit hospitals, philanthropy and health insurers.

NACTN has combined support from multiple sources to develop a clinical trials network.

We believe that consortia of funding sources are required to support clinical trials of new therapy in SCI.

Randomized multicenter clinical trials require extensive planning that frequently takes 1-2 years to complete. Additional time is required for harmonizing the protocol with specific requirements that individual IRBs request to create a master protocol that is acceptable to all of the IRBs,a process that requires frequent protocol revisions and IRB submissions.

The complexity of planning and initiating multicenter trials and the cost of such trials have been major factors in preventing new therapies from being brought from the laboratory into clinical practice.

The most important elements in building a clinical trials network are the dedication of the clinical staff and the willing participation of patients, to whom we owe our gratitude

High Performance Liquid Chromatographic Assay for Riluzole in Rat Plasma, Brain, Spinal Cord and Liver

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Abstract:

A simple method was developed and validated for the quantification of riluzole in rat plasma, brain, spinal cord and liver. Separation was on a C18 reversed-phase column with an UV detection at 263nm. Liquid-liquid extraction with ethyl acetate was used for the extraction of riluzole from these biological samples. The assay was linear from 7.8 to 1000ng/ml, with the lower limit of quantification (LLOQ) of 7.8ng/ml for rat plasma, and linear from 31.25 to 4,000ng/g, with LLOQ of 31.25ng/g for rat brain, spinal cord and liver. The overall mean recoveries of riluzole from rat plasma, brain, spinal cord and liver samples were 96.84±3.73%, 99.96±5.75%, 103.60±6.87% and 95.95±9.42% (n=27), respectively. The accuracies and precisions for these samples were within 90-110% and less than 4%. The assay provided good reproducibility and can be used to support the studies of impacts of acute spinal cord injury on riluzole pharmacokinetics in rat model.

Keyword: *Riluzole; HPLC; Rat Plasma; Rat Brain; Rat Spinal Cord; Rat Liver*

Abbreviation: ALS, amyotrophic lateral sclerosis; ASCI, acute spinal cord injury; SCs, spinal cords;

1. Introduction

Riluzole (Figure 1), a voltage-dependent sodium channel blocker with anti-glutamatergic activity, shows neuroprotective effect and promotes functional and neurological recoveries in animal models of brain and spinal cord ischemic and traumatic injury [1-4]. However, major adverse effects have been reported following the administration of riluzole including psychiatric disturbances and liver damage [5,6].

Acute spinal cord injury (ASCI) results in a devastating loss of neurological function below the level of injury and adversely affects physiological situations of multiple systems within the body (such as microvascular blood flow in the liver, spleen and gastrointestinal tract), and may change the pharmacokinetic behaviors of drugs. However, it is unclear how these alterations will affect the kinetics of therapeutic drug absorption, distribution, metabolism and excretion. It has been reported that the pharmacokinetics of several drugs, such as paracetamol [7], theophylline [8], dantrolene [9], aminoglycosides [10,11], and lorazepam [12] are significantly altered in SCI patients in comparison with able-bodied subjects. Clinical reports on drug kinetics in SCI are often anecdotal, because it is extremely difficult to perform systematic pharmacokinetic studies in SCI patients. This difficulty is due to the important interindividual variability in injury extent and location. Therefore, the use of experimental models appears to be a suitable strategy for understanding pharmacokinetic alterations due to ASCI, as well as the pathophysiological mechanisms involved [13,14].

By comparing riluzole pharmacokinetic behaviors in plasma, brain, spinal cord, and liver in a spinal cord injured rat model with those in uninjured control group, we can begin to understand the impacts of ASCI on riluzole pharmacokinetics. A new and simple method for the quantification of riluzole in rat plasma, brain, spinal cord and liver is a pre-requisite to support these studies. Analysis based on high performance liquid chromatography with ultraviolet detection (HPLC-UV) [15,16] or coupled with tandem mass spectrometry (LC/MS/MS) [17] have been described for the determination of riluzole in human plasma and urine. However, they do not appear to be able to quantitate the drug in CNS tissues and liver of animals. On the other hand, although some simple HPLC-UV methods [18,19] have been published for quantifying riluzole in rat brain, mouse plasma and central nervous system tissues, however, there

was no method to study the uptake of riluzole in rat liver. Moreover, if different methods (HPLC conditions and biological samples pre-treatment) are used for quantification and preparation of riluzole in different tissues and organs, it will cause inconveniences and errors. Therefore, we establish a rapid and sensitive HPLC method for the quantification of riluzole in rat plasma, brain, spinal cord and liver, capable of satisfying our studies and detection requirements.

2. Experimental

2.1. Materials and reagents

Riluzole and 5-methoxypsoralen (5-MOP, internal standard; I.S., Figure 1) were purchased from Sigma-Aldrich (U.S.A). 2-Hydroxypropyl-β-cyclodextrin (HP-β-CD) was purchased from Sigma-Aldrich (U.S.A). All the solvents and chemicals were of HPLC or analytical grade. Methanol, acetonitrile and acetic acid (glacial) were obtained from EMD (U.S.A.). Ethyl acetate and ammonium acetate were purchased from J.T. Baker and Fisher Scientific (U.S.A.), respectively.

Stock solutions were prepared by dissolving riluzole and the internal standard in methanol at the concentration of 1mg/ml. Working standard solutions of rilzuole(100μg/ml) and I.S. (50ug/ml and 10ug/ml) were prepared from the stock solutions by the dilution with methanol and double distilled water (1:1, v/v), and kept at +4°C in 15ml plastic tubes.

Drug-free plasma, brain, spinal cord and liver tissues were obtained from female Wistar rats and stored at -80°C after collection and processing.

2.2. Chromatographic apparatus and conditions

The HPLC assay was developed using Waters system equipped with 717 plus auto sampler, 515 HPLC pump and 2996 UV detector set at 263 nm. Baseline resolution was achieved on Waters Symmetry® C18 column (3.0×150 mm, 3.5μm) with Symmetry® C18 guard column (2.1×10 mm, 3.5μm), eluted at the flow rate of 0.45ml/min, with the mobile phase of acetonitrile: methanol: 0.1 M ammonium acetate (3:2:5, v/v/v), adjusted with acetic acid to pH 6.5.

2.3. Sample preparations

Plasma Samples: One hundred μL plasma was mixed with 10μl of 5-MOP (50μg/ml, internal standard). After the addition of 500 μl of ethyl acetate, the mixture was vortexed for 30s and then centrifuged at 16,000 g for 20 minutes. Of the clear organic layer, about 500 μl was withdrawn and evaporated to dryness under the air stream. The residue was reconstituted in 1 ml of mobile phase, then mixed on a vortex for 30s and centrifuged at 16,000 g for 20 minutes. The clear supernatant samples were transferred into auto-sampler vials.

Brain, spinal cord and liver samples: The brain, spinal cord (cervical and thoracic segments) and liver samples were thawed, accurately weighed, and added to appropriate volumes of normal saline with the weight/volume ratio of 0.2. The samples were vortex-mixed and homogenized with tissue tearor homogenizer until thoroughly homogenized. 100μl brain, spinal cord (cervical and thoracic segments) and liver homogenate sample were mixed with 10μl of 5-MOP (50μg/ml, internal standard), respectively. After the addition of 200μl ethyl acetate, the mixture was vortexed for 30s and then centrifuged at 16,000 g for 20 minutes. Of the clear organic layer, about 200μl was withdrawn and evaporated to dryness under the air stream. The residue was reconstituted in 1 ml of mobile phase, then mixed on a vortex mixer for 30s and centrifuged at 16,000 g for 20 minutes. The clear supernatant samples were transferred into auto-sampler vials.

2.4. Method validation

2.4.1. Recovery

For the rat plasma samples, the recovery and reproducibility of the extractions of both riluzole and 5-MOP were determined by the analysis of nine independent spiked samples at each of three quantification levels (15.6, 125 and 1,000ng/ml). Results were calculated by comparing the peak areas obtained from the direct injections of standard

solutions of compounds with those by extractions from spiked rat plasma samples through the assay procedure previously described.
For the rat brain, spinal cord and liver samples, the recovery and reproducibility of the extractions of both riluzole and 5-MOP were determined by the analysis of nine independent fortified samples at three quantification levels (31.25, 500 and 4,000ng/g). Results were calculated by comparing the peak areas obtained from the direct injections of standard solutions of compounds with those by extractions from spiked rat brain, spinal cord and liver samples through the assay procedure described.

2.4.2. Linearity, accuracy and precision

Calibration curves were prepared according to the method described in section 2.3, except after dryness, the residue was reconstituted in 100µl of mobile phase for rat plasma, brain, spinal cord and liver samples, then mixed on a vortex for 30s and centrifuged at 16,000 g for 20 minutes. The clear supernatant samples were transferred into auto-sampler vials with 150µl glass inserts. A least-squares linear regression method ($1/x^2$ weighting) was used to determine the slope, intercept and correlation coefficient of linear regression equation. The lower limit of quantification (LLOQ) was determined based on the signal-to-noise ratio of 10:1.
For the rat plasma samples, the linearity of response was evaluated in the concentration range of 7.8 to 1,000ng/ml. Spiked plasma samples at levels of 7.8, 15.6, 31.25, 62.5, 125, 250, 500 and 1,000ng/ml were used as daily calibrators. Three spiked plasma samples at each level of 15.6, 125 and 1,000ng/ml were analyzed on the same day to calculate the accuracy and intra-day precision of the assay, using the established calibration curve. Inter-day precisions at the three levels were determined on three separate days.
For the rat brain, spinal cord and liver samples, the linearity of response was also evaluated between 31.25 and 4,000ng/g. Spiked brain, spinal cord and liver samples at levels of 31.25, 62.5, 125, 250, 500, 1,000, 2,000 and 4,000ng/g were used as daily calibrators. Triplicate spiked CSF samples at concentrations of 31.25, 500 and 4,000ng/g were prepared and analyzed on the same day to calculate the accuracy and

intra-day precision of the assay, using the established calibration curve. Inter-day precisions at the three levels were determined on three separate days.

2.4.3 Stability

Short-term (room temperature for 8h), post-processing (4°C for 24h), long-term (-80°C for one month) and three freeze-thaw cycle stabilities were determined.

2.5. Pharmacokinetic study in vivo

This part is a collaborative effort with the group of Dr. Michael Fehlings (Professor of Neurosurgery), Division of Genetics & Development, Toronto Western Research Institute (TWRI) in Canada.

2.5.1 Animals

Normal female Wistar rats (body weight between 250 and 300 g) were used and kept in an environmentally controlled room.

2.5.2 Experimental design

Riluzole (Sigma, R116) was solubilized in 22.5% 2-hydroxypropyl-β-cyclodextrin (HB-β-CD, Sigma) to a concentration of 6 mg/ml, and then diluted with normal saline to a concentration of 2.4 mg/ml for injections. One regimen group was administrated with riluzole at a single dose of 8 mg/kg by intraperitoneal injection, plasma, brain, spinal cord (cervical and thoracic segments) and liver were collected at 6hr and 9hr, respectively (n=4 for each time point). The plasma samples of 500μl were collected by adding 100μl of 50mM EDTA to 1ml of whole blood and centrifuging at 3,000 g for 10 min at 4^{o}C. All the plasma, brain, spinal cord and liver samples were shipped in packages with dry ice, from University of Toronto to College of Pharmacy, University of Houston, and were stored at -80° C until analysis.

3. Results and discussion

3.1 HPLC chromatogram

Riluzole was recovered efficiently from the rat plasma, brain, spinal cord and liver using ethyl acetate. Retention times of Riluzole and I.S. were 6.9 and 8.9 minutes, respectively. No interfering peaks were observed in chromatograms of these blank biological matrixes (Figure 2).

3.2 Recovery, Accuracy and Precision

Plasma samples: The LLOQ was 7.8ng/ml and the assay was linear from 7.8 to 1,000ng/ml, with correlation coefficients of 0.9996. In plasma samples, intra-day and inter-day precision coefficients of variation at each riluzole level of 15.6, 125 and 1,000ng/ml were within 3%, the accuracies were 103.38%, 95.99% and 94.56% (n=9), respectively. The recoveries were 92.69-98.94% with a mean of 96.84±3.73% (n=27) (Table 1).

Brain, spinal cord and liver samples: The LLOQ was 31.25ng/g and the assay was linear from 31.25 to 4,000ng/g, with correlation coefficients of 0.9991, 0.9999 and 0.9990 for rat brain, spinal cord and liver samples. The intra-day and inter-day precision coefficients of variation at each riluzole level of 62.5, 500 and 4,000ng/g were within 4% for these biological matrixes, the accuracies were all within 90-110% which satisfied the requirements of FDA (n=9 at each concentration). The recoveries were 96.10-107.62%, 98.06-112.50% and 87.55-108.31% with a mean of 99.96±5.75%, 103.60±6.87% and 95.95±9.42% (n=27) for brain, spinal cord and liver samples, respectively (Table 1)

3.3 Stability

The stability of riluzole in rat plasma, brain, spinal cord and liver were evaluated by analyzing three replicates of quality control samples at three different concentrations after short-term (room temperature, 8h), post processing (4°C for 24h), long-term cold storage (-80°C, one month) and within three freeze-thaw cycles. All the samples were stable and displayed 85-115% recoveries after various stability tests.

3.4 Pharmacokinetic studies

The validated analytical method was employed to study the pharmacokinetic behaviors of riluzole in rats. The riluzole concentrations in cervical and thoracic spinal cords were highest and comparable, and could be considered as within the same compartment pharmacokinetically, after I.P. administration of single dose of the cyclodextrin-enclosed riluzole liquid formulation. The riluzole concentration in liver was no statistically different compared to those of spinal cords, which was in the same level. The riluzole concentration in the brain yielded a significantly lower levels than those in the spinal cord and liver (almost half of those in SCs). The brain may be considered a distinct compartment from that of spinal cords and liver. The plasma concentration of riluzole was significantly lower than those in brain, liver and spinal cords. A representative chromatogram of rat liver, removed 6hr after I.P. administration of riluzole, is showed in Figure 2. The concentrations of riluzole in these biological matrixes were represented in Figure 3.

4. Conclusion

A specific, accurate and precise HPLC method with a UV detection was developed and validated for the quantification of riluzole in rat plasma, brain,spinal cord and liver.

The HPLC-UV method developed by Milena Colovic et al. [19] for quantifying the riluzole in mouse plasma and central nervous system tissues used the solid-phase extraction, which was not as convenient as liquid-liquid extraction with quantities of samples. Adriana Maltese et al. [18] developed the HPLC-UV method only for quantification of riluzole in rat brain. Our method enlarged the range of pharmacokinetic applications, including rat plasma, brain, spinal cord and liver. The method we developed has LLOQ of 7.8ng/ml for plasma samples and 31.25ng/g for other biological matrixes, and uses liquid-liquid extraction with coefficients of variation of < 4% for all the samples. Therefore, the assay can be used to investigate the impacts of acute spinal cord injury on riluzole pharmacokinetics, such as absorption, distribution, metabolism and elimination.

5. Acknowledgement

This research was supported by North American Clinical Trial Network (NACTN), Christopher Reeve Foundation. The animal work was done by Toronto Western Research Institute (TWRI), Toronto Western Hospital, University Health Network, University of Toronto.

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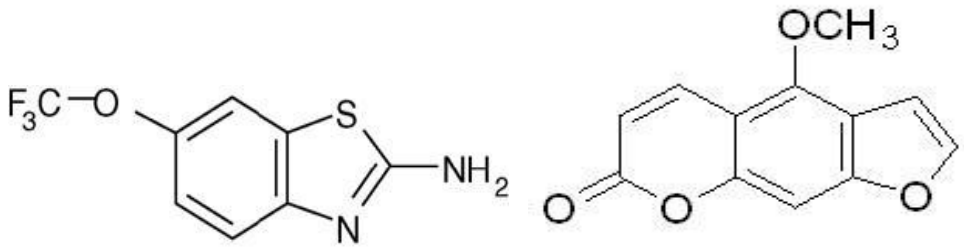
Figure Captions

Figure 1. Chemical Structures of Riluzole and 5-Methoxypsoralen.

Figure 2. Authentic HPLC Chromatograms of Extracts of (A) Blank Liver, (B) Spiked Liver Sample at LLOQ of 31.25ng/g, (C) Spiked Liver Sample at 4,000ng/g, (D) Liver Removed 6hr After Single I.P. Administration (8mg/kg). (Retention time: 8.9 min for riluzole and 6.9 min for I.S., respectively)

Figure 3. Mean Plasma, Brain, Spinal cord (cervical and thoracic segments) and Liver Concentrations of Riluzole in Female Wistar Rats at 6hr and 9hr after Single Intraperitoneal Injection (8mg/kg). Each Point Represents the Mean ± SD of Four Animals.



Riluzole 5-MOP

Figure 1.

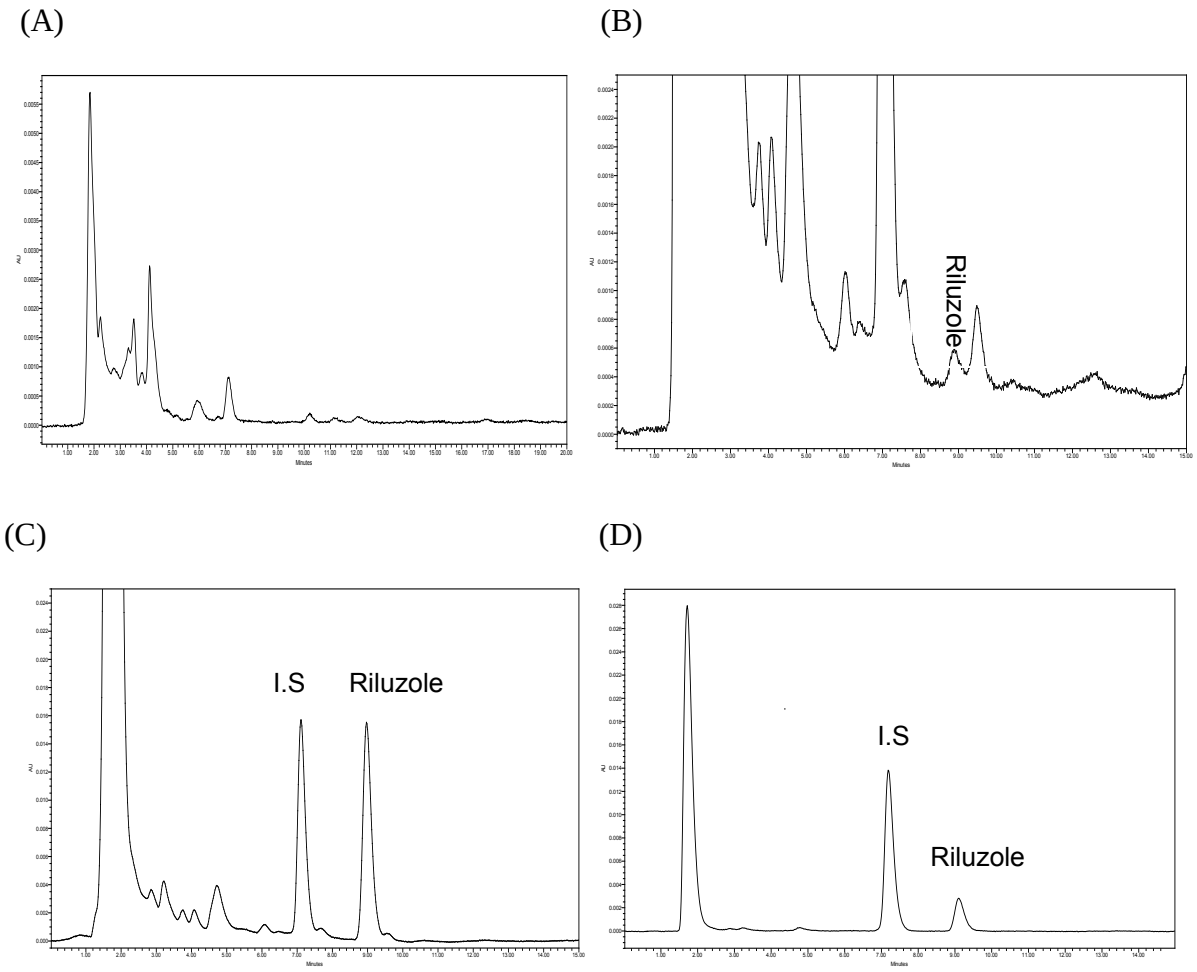


Figure 2.

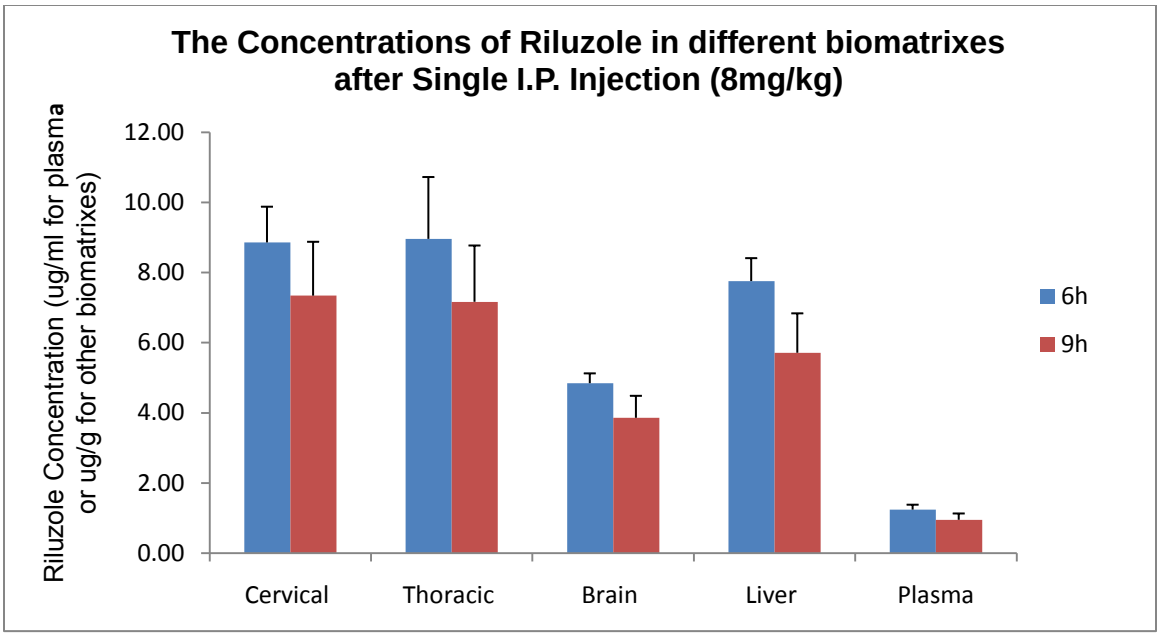

The Concentrations of Riluzole in different biomatrixes after Single I.P. Injection (8mg/kg)
Riluzole Concentration (ug/ml for plasma or ug/g for other biomatrixes)
12.00
10.00
8.00
6.00
4.00
2.00
0.00
Cervical
Thoracic
Brain
Liver
Plasma
6h
9h

Figure 3.

.

Table 1.

Accuracy, Precision and Recovery of Riluzole at Levels of 15.6, 125 and 1,000ng/ml for Rat Plasma Samples and at Levels of 62.5, 500 and 4,000ng/g for Rat Brain, Spinal Cord and Liver Samples (n=9 each).

Added Concentration (ng/ml for plasma and ng/g for others)	Measured Concentrations (ng/ml) mean ± SD	Accuracy(%)	Precision (%)		Recovery(%) mean ± SD
			Intra-day	Inter-day	
Plasma					
15.6	16.13 ± 0.26	103.38	1.3	1.6	92.69 ± 2.42
125	119.98 ± 1.81	95.99	0.8	1.5	98.91 ± 1.57
1,000	945.63 ± 26.24	94.56	1.1	2.8	98.94 ± 2.76
Brain					
62.5	62.33 ± 0.85	99.88	1.4	1.4	107.62 ± 2.03
500	490.05 ± 8.32	98.01	0.3	1.7	96.17 ± 1.69
4,000	3947.38 ± 46.61	98.68	0.2	1.2	96.10 ± 1.14
Spinal cord					
62.5	57.94 ± 1.50	92.70	0.4	2.6	112.50 ± 3.58
500	506.68 ± 6.17	101.34	2.0	1.2	100.23 ± 1.25
4,000	4048.62 ± 66.72	101.22	0.2	1.6	98.06 ± 1.62
Liver					
62.5	59.35 ± 1.35	94.95	1.4	2.3	108.31 ± 2.99
500	500.63 ± 6.98	100.13	0.3	1.4	91.98 ± 1.31

4,000	3893.48 ± 130.78	97.34	0.2	3.4	87.55 ± 2.95

High Performance Liquid Chromatographic Assay for Riluzole in Human Plasma and Cerebrospinal Fluid (CSF) Samples

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Abstract:

A specific, accurate and precise HPLC assay was developed and validated for the quantification of riluzole in a small volumes (200µl) of human plasma and CSF. Separation was on a C18 reversed-phase column with UV detection at 263nm. Liquid-liquid extraction with ethyl acetate and solid phase extraction were used for the extraction of riluzole from the human plasma and CSF samples, respectively. The assay was linear from 7.8 to 1000 ng/ml, with a lower limit of quantification (LLOQ) of 7.8ng/ml. The overall mean recoveries of riluzole from human plasma and CSF samples were 78.8±7.2% (n=27) and 71.1±7.5% (n=36), respectively. The accuracy and precision for human plasma samples were within 93-107% and less than 9%, respectively; for human CSF samples, the accuracy and precision were within 97-101% and less than 13%, respectively. The assay is a prerequisite for the pharmacokinetic and pharmacodynamic evaluation of riluzole as a neuroprotective therapy in an ongoing clinical trial of patients with acute spinal cord injury (ASCI).

Keyword: *Riluzole; HPLC; Human plasma; Human CSF, SCI*

Abbreviation: ALS, amyotrophic lateral sclerosis; SCI, spinal cord injury; CSF, cerebrospinal fluid; LLOQ, lower limit of quantification; LP, low CSF protein; IP, intermediate CSF protein; HP, high CSF protein; ASIA, American spinal injury association;

1. **Introduction**

Riluzole (Figure 1), a voltage-dependent sodium channel blocker with anti-glutamatergic activity, has been approved by FDA for the treatment of patients with amyotrophic lateral sclerosis (ALS) for almost 10 years. In ALS, riluzole treatment has prolonged patients' lives by 2-3 months [1, 2].

Riluzole has also been investigated in a wide variety of experimental models for acute neurodegerative diseases[3], including spinal cord injury (SCI). A neuroprotective effect of riluzole has been demonstrated in animal models of brain and spinal cord ischemia and traumatic injury [3-5]. The neuroprotective effect of riluzole appears to be due to the blockade of voltage-sensitive sodium channels whose persistent activation following injury results in entry of calcium and sodium into neurons, glia and vascular cells leading to cell death [6]. Antagonism of presynaptic calcium-dependent glutamate release may also be a mechanism [7].

The North American Clinical Trial Network (NACTN), a consortium of nine university affiliated hospitals is currently carrying out a Phase I clinical trial of riluzole as a treatment for SCI [8]. A robust method for the quantification of riluzole in human plasma and CSF is a prerequisite to support the clinical studies. Methods have been described for the determination of riluzole in human plasma and urine based on high performance liquid chromatography with ultraviolet detection (HPLC-UV) [9, 10] or coupled with tandem mass spectrometry (LC/MS/MS) [11]. There was no method described to study

the uptake of riluzole in human CSF. Therefore, we have established a rapid and sensitive HPLC method for the quantification of riluzole in human plasma and CSF.

2. Experimental

2.1. Materials and reagents

Riluzole and 5-methoxypsoralen (5-MOP, internal standard; I.S., Figure 1) were purchased from Sigma-Aldrich (U.S.A). All the solvents and chemicals were of HPLC or analytical grade. Methanol, acetonitrile and acetic acid (glacial) were obtained from EMD (U.S.A.). Ethyl acetate and ammonium acetate were purchased from J.T. Baker and Fisher Scientific (U.S.A.), respectively.

Stock solutions were prepared by dissolving riluzole and the internal standard, respectively, in methanol at the concentration of 1mg/ml. Working standard solutions (10μg/ml) were prepared from the stock solutions by a dilution with methanol and double distilled water (1:1, v/v), and kept at +4 °C in 15ml plastic tubes.
Blank, drug-free human plasma and CSF used for the preparation of spiked samples were obtained from the Methodist Hospital (Houston, TX, U.S.A.) and frozen at −80°C until use. Human plasma samples (50ml) were collected in sodium heparin-treated tubes. Twenty-six of CSF samples (volume range of 1 to 14ml) were received, with CSF protein concentrations ranging from 22 to 1,345mg/dl, and grouped into low CSF protein (LP) and high CSF protein (HP) groups. Pooled LP sample of 47ml has the final protein

concentration of 53.9mg/dl, and HP of 41.5ml with protein concentratoin of 348.2mg/dl; Intermediate CSF protein (IP) group of 10ml was prepared by mixing LP and HP samples at a ratio of 7:3 (v/v) to yield the protein concentration of 142.2mg/dl.

2.2. Chromatographic apparatus and conditions

The HPLC assay was developed using Waters system equipped with 717 plus auto sampler, 515 HPLC pump and 2996 UV detector set at 263 nm. Baseline resolution was achieved on Waters Symmetry® C18 column (3.0×150 mm, 3.5µm) with Symmetry® C18 guard column (2.1×10 mm, 3.5µm), eluted at the flow rate of 0.45ml/min, with the mobile phase of acetonitrile: methanol: 0.1 M ammonium acetate (3:2:5, v/v/v), adjusted with acetic acid to pH 6.5.

2.3. Sample preparations

Two hundred µL plasma was mixed with 10µl 5-MOP (10µg/ml, I.S). After addition of 1ml ethyl acetate, the mixture was vortexed for 30s and then centrifuged at 15,000 rpm for 20 minutes. Of the clear organic supernatant, 1ml was taken and evaporated to dryness under a stream of air. The residual was reconstituted in 200µl mobile phase, then vortexed for 30s and centrifuged at 15,000 rpm for 20 minutes. The clear supernatant was transferred into auto-sampler vials with 150µl glass inserts.
Two hundred µL CSF was mixed with 10µl 5-MOP (10µg/ml, I.S). The mixed aliquot was applied for solid phase extraction onto 3ml Octadecyl (C18) speedisk columns

(J.T.Baker), previously activated with 2ml methanol and 2ml water. After washed by 1ml water and then eluted with 1ml methanol, the methanol eluent was evaporated to dryness under a stream of air. The residual was reconstituted in 200μl mobile phase, then vortexed for 30s and centrifuged at 15,000 rpm for 20 minutes. The clear supernatant was transferred into auto-sampler vials with 150μl glass inserts.

2.4 The effects of different CSF protein concentrations on recovery and accuracy of the quantification of CSF samples

The blank CSF samples received contained various CSF protein concentrations ranging from 22 to 1,345mg/dl. Riluzole is highly protein binding drug, 96% in plasma. Therefore, we investigated if such different CSF protein concentrations will have impacts on the quantification of CSF samples. As above mentioned, we classified twenty-six CSF samples into three groups [LP (53.9mg/dl), IP (142.2mg/dl) and HP (348.2mg/dl)]. Spiked human CSF samples of these three groups were prepared at riluzole levels of 7.8, 15.6, 31.25, 62.5, 125, 250, 500 and 1,000ng/ml as daily calibrators. Calibration curves for LP, IP and HP samples were run five, two and five times, respectively. Three spiked CSF samples at each riluzole concentrations of 62.5 and 1,000ng/ml were prepared at each CSF protein level and analyzed on the same day to calculate the accuracies. The procedure was repeated on a second day.

2.5. Method validation

2.5.1. Recovery

For the human plasma samples, the recovery and reproducibility of the extractions of both riluzole and 5-MOP were determined by the analysis of nine independent spiked samples at each of three quantification levels (15.6, 125 and 1,000ng/ml). Results were calculated by comparing the peak areas obtained from the direct injections of standard solutions of compounds with those by extractions from spiked human plasma samples through the assay procedure previously described.

For the human CSF samples, the recovery and reproducibility of the extractions of both riluzole and 5-MOP were determined by the analysis of eighteen independent fortified samples at two quantification levels (62.5 and 1,000ng/ml). Results were calculated by comparing the peak areas obtained from the direct injections of standard solutions of compounds with those by extractions from spiked human CSF samples through the assay procedure described.

2.5.2. Linearity, accuracy and precision

The lower limit of quantification (LLOQ) was defined as the mean riluzole concentration resulting in a peak response with a signal to noise ratio of 10.

For the human plasma samples, the linearity of response was evaluated in the concentration range of 7.8 to 1,000ng/ml. Spiked plasma samples at levels of 7.8, 15.6, 31.25, 62.5, 125, 250, 500 and 1,000 ng/ml were used as daily calibrators. Three spiked

plasma samples at each level of 15.6, 125 and 1,000ng/ml were analyzed on the same day to calculate the accuracy and intra-day precision of the assay, using the established calibration curve. Inter-day precisions at the three levels were determined on three separate days.

For the human CSF samples, the linearity of response was also evaluated between 7.8 and 1,000ng/ml. Triplicate spiked CSF samples at concentrations of 62.5 and 1,000ng/ml were prepared at individual CSF protein levels (LP, IP and HP) and analyzed on the same day to calculate the accuracy and intra-day precision of the assay, using the individual calibration curves and the pooled calibration curve, respectively. Inter-day precisions at 62.5 and 1,000ng/ml were determined on two separate days.

2.6. Stability test

We investigated three kinds of stabilities for human plasma and CSF samples, bench-top stability, freezing and thawing stability and long-term stability.

The bench-top stability of human plasma samples at 15.6, 250 and 1,000ng/ml levels was investigated by repeated injection immediately after work-up and 8 and 24 h post the preparation. The bench-top stability of human CSF samples was investigated at 15.6, 125 and 1,000ng/ml levels. The influence of temperature change on the stored human plasma and CSF samples was evaluated by analyzing samples (n = 3) of three

riluzole levels after three freezing and thawing cycles (−80°C), and being stored at +4°C for 48h. The long-term stability of the riluzole in spiked samples was evaluated after storage at –80°C for one month at respective three levels for human plasma and CSF.

2.7. Application to clinical studies

The North American Clinical Trial Network (NACTN) is currently engaged in a Phase I clinical trial for the treatment of SCI [8]. The described method has been employed in the clinical studies to quantify riluzole concentrations in plasma and CSF in support of pharmacokinetic evaluations. Patients received riluzole 50 mg orally or by nasogastric tube twice daily for 14 days. On the 3^{rd} and 14^{th} days, blood samples (2ml) were taken before the riluzole dose for the trough and 2hr post dose for the peak concentrations, respectively. Plasma was separated by centrifugation at 2,700 g for 10 minutes immediately after collection, and frozen at -80°C until analysis. The CSF withdrawal is indicated only if there is a suspicion of meningitis or if there is a need for myelography.

3. Results and discussion

3.1 HPLC chromatogram

Riluzole was recovered efficiently from the human plasma and CSF using ethyl acetate and solid phase extraction, respectively. Retention times of Riluzole and I.S. were 6.9

and 8.9 minutes, respectively. No interfering peaks were observed in chromatograms of the blank plasma and CSF (Figure 2).

3.2 Recovery, Accuracy and Precision

The LLOQ was 7.8ng/ml and the assay was linear from 7.8 to 1,000ng/ml, for both plasma and CSF samples with correlation coefficients of 0.9995 and 1, respectively. In plasma samples, intra-day and inter-day precision coefficients of variation at each riluzole level of 15.6, 125 and 1,000ng/ml were within 9%. The accuracies were 106.5%, 93.8% and 102.6% (n=9), respectively. The recoveries were76.7-82.4% with a mean of 78.8±7.2% (n=27) (Table 1). For CSF samples, the intra-day and inter-day precision coefficients of variation of two concentrations (62.5ng/ml and 1,000ng/ml) were within 13%. The accuracies were 97.6% and 100.6% (n=18) and the recoveries were 68.7-73.6% with a mean of 71.1±7.5% (n=36) (Table 1).

3.3 Stability

The bench-top stability of pretreated samples, the temperature influence on the stored samples and long-term stability, expressed as percentage remaining of the original concentration, were summarized in Tables 2. Both human plasma and CSF samples of riluzole were stable for longer than 24h at room temperature, and longer than 48h at +4°C. Furthermore, riluzole was stable up to one month when stored at -80°C.

Therefore, all the clinical samples can be stored at -80°C until HPLC assay within a month.

3.4 The effects of CSF protein on recovery and accuracy of the quantification of CSF samples

The different CSF protein concentrations had no significant effects on the recovery and accuracy of the quantification of CSF samples (Tables 3 & 4). Therefore, the pooled calibration curve will be used as daily calibrator for the future experiments.

3.5 Clinical Trial

Riluzole was quantified with triplicate measurements in the plasma samples from a patient (Age: 53 years; ASIA A) who enrolled in Phase I clinical trial of riluzole for the treatment of SCI. The individual plasma peak and trough concentrations of riluzole on the 3rd (Figure 3) and 14th day of the 14-day treatment were measured.

4. Conclusion

A specific, accurate and precise HPLC method with UV detection was developed and validated for the quantification of riluzole in human plasma and CSF. The method requires only a small volume (200µl) of plasma or CSF, and the assay sensitivity at the concentration of 7.8ng/ml is sufficient for clinical studies.

The HPLC-UV method developed by H. J. M. van Kan et al. [9] needed a large volume of human plasma (500µl) and LLOQ was 20ng/ml for the determination of riluzole in human plasma or serum. Le Liboux et al. developed HPLC-UV [10] and LC/MS/MS methods [11] for the quantifications of riluzole in human plasma and urine. The HPLC method also required a large volume of human plasma (1ml), and the solid-phase extraction was less convenient than liquid-liquid extraction. The LLOQ of 0.5ng/ml was achieved with the LC/MS/MS method; however, the inter-day precision is large, ranging from 11% to 20%. Our method has an LLOQ of 7.8ng/ml and uses liquid-liquid extraction with coefficients of variation of < 9% for human plasma samples.

To our knowledge, the present paper is the first publication in which a quantitative assay for riluzole in human CSF is fully described. The solid-phase extraction is adopted for CSF samples, with an LLOQ of 7.8ng/ml and coefficients of variation of < 13%. Our results demonstrated that the CSF protein (22-1,345mg/dl) has no effects on the accuracy of riluzole quantification.

The present assay can be employed to quantify riluzole levels in human plasma and CSF over a wide range of 7.8-1,000ng/ml.

In patients with ALS riluzole serum concentrations are in the range of 10-500ng/ml with a standard drug regimen of 50 mg twice daily [10, 11]. The present assay has been

successfully employed for the quantification of riluzole in blood plasma in nine SCI patients who have been treated with a dosing regimen of 50 mg orally twice daily.

5. Acknowledgement

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Figure Captions

Figure 1. Chemical Structures of Riluzole and 5-Methoxypsoralen.

Figure 2. Authentic HPLC Chromatograms of (A) Extract from Blank Human Plasma, (B) Spiked Plasma Sample, (C) Extract from Blank Human CSF, (D) Spiked CSF Sample (Riluzle:1,000 ng/ml, retention time of 8.9min; I.S. : 500 ng/ml, retention time of 6.9min).

Figure 3. HPLC Chromatograms of (A) Extract from Blank Patient Plasma, (B) Patient Plasma Sample (Peak sample of 3rd day treatment of oral riluzole, 50mg twice daily for 14 days).

Table 1.

Accuracy, Precision and Recovery of Riluzole at Levels of 15.6, 125 and 1,000ng/ml for Human Plasma Samples (n=9 each) and at Two Concentrations (62.5ng/ml and 1,000ng/ml) Levels for Human CSF Samples (n=18).

Added Concentration (ng/ml)	Measured Concentrations (ng/ml) mean ± SD	Accuracy(%)	Precision (%)		Recovery(%) mean ± SD
			Intra-day	Inter-day	
Plasma (n=9 each)					
15.6	16.6 ± 0.7	106.5	3.2	3.9	76.7 ± 10.7
125	117.2 ± 3.9	93.8	8.3	3.3	77.2 ± 4.2
1000	1025.7 ± 24.3	102.6	3.9	2.4	82.4 ± 3.5
CSF (n=18 each)					
62.5	61.0 ± 3.7	97.6	4.4	6.8	68.7 ± 4.6
1000	1005.9 ± 123.0	100.6	3.8	12.3	73.6 ± 9.1

Table 2.

Stability (%Remaining) of Riluzole in human plasma and CSF samples, at room temperature, after repeated freeze-thaw and long-term storage.

	Short-term stability				Long-term stability
Added Riluzole Concentration (ng/ml)	Room Temperature 8h	Room Temperature 24h	Stored at +4°C for 48h	Freeze and Thaw_three times	Stored at -80°C for one month
Plasma					
15.6	94.5	99.7	99.1	91.4	98.5
250	102.6	101.4	103.9	103.5	102.4
1000	101.1	101.7	102.0	102.1	100.5
CSF					
15.6	96.1	113.0	107.1	112.2	99.2
125	95.6	95.8	97.0	106.0	98.1
1000	102.5	102.3	88.8	104.3	98.4

Table 3.

Calibration Curves of Riluole with CSF of Different Protein Concentrations [LP Samples (53.9mg/dl), IP Samples (142.2mg/dl) and HP Samples (348.2mg/dl)].

Groups	N	Slope of Standard Curve	Mean ± SD	CV(%)
LP	5	0.00137	0.00138 ± 0.00023	16.6
		0.00163		
		0.00158		
		0.00116		
		0.00114		
IP	2	0.00133	0.00128 ± 0.00007	5.7
		0.00123		
HP	5	0.00133	0.00137 ± 0.00009	6.4
		0.00137		
		0.00149		
		0.00142		
		0.00126		
Pooled Standard Curves	12		0.00137 ± 0.00015	11.2

Table 4.

Comparison of Accuracies and Precisions Using Individual and Pooled Calibration Curves.

Added Riluzole Concentration(ng/ml)	CSF Protein Groups	Calculation using Respective Curves (n=6) Accuracy (%)	Precision (%)	Calculation using Pooled Curves (n=18) Accuracy (%)	Precision (%)
62.5	LP	89.4	9.7	97.6	6.0
	IP	103.7	2.9		
	HP	99.8	5.6		
1000	LP	90.6	20.6	100.6	12.2
	IP	110.2	3.5		
	HP	103.7	5.5		

Table 5.

The Quantification of Riluzole in Acute Spinal Cord Injured Patient who Enrolled in Phase I Clinical Trial

Date	C_{peak} (ng/ml)		C_{trough} (ng/ml)	
	Individual Conc.	Mean ± SD (CV%)	Individual Conc.	Mean ± SD (CV%)
Day 3	115.8 116.0 115.7	115.8 ± 0.1 (0.1)	20.9 18.9 21.1	20.3± 1.2 (6.0)
Day 14	38.6 33.7 34.7	35.7 ± 2.6 (7.3)	17.1 15.5 15.4	16.0 ± 1.0 (6.0)

Graded Redefined Assessment of Sensibility Strength and Prehension (GRASSP): Psychometric Development of an Upper Limb Impairment Measure for Individuals with Traumatic Tetraplegia

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Background

• With the advent of new interventions targeted at both acute and chronic spinal cord injury (SCI) it is critical that improved techniques and protocols to evaluate changes in upper limb impairment/function be developed

• The GRASSP is a multi-modal (sub-tests) quantitative clinical test developed by an international collaboration. The GRASSP was developed to be responsive to change, assess the extent of natural recovery over the acute to chronic phases, and evaluate the effect of novel interventions. Impaired upper limb function is primarily determined by sensory and motor deficit. Absent or decreased upper limb function is one of the major disabilities affecting decreased functional independence, productivity, and quality of life for individuals with tetraplegia[1]

• Identifying the deficits, impairments and pattern(s) of recovery is the initial step in future development of therapeutic rehabilitative interventions. The increasing incidence of incomplete tetraplegia results in more variable sensory, motor impairments and functional consequences making methods for quantification of impairment a priority

• OBJECTIVES:

- To define a scoring approach for GRASSP to best represent sensorimotor impairment of the upper limb in tetraplegia
- To establish the psychometric properties of inter rater, test retest reliability and construct validity
- To examine the relationships between impairment and function of the upper limb

Methods & Materials

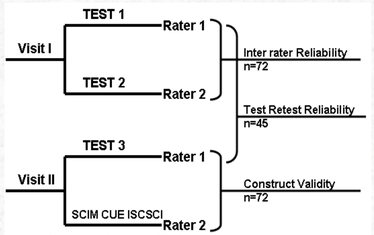
• A cross-sectional (n=72) multi-centre (n=7) study was conducted in North America and Europe. Study participants included in the trial presented with chronic traumatic tetraplegia (complete/incomplete), severity of injury between Occiput to T1, neurologically stable and able to provide informed consent

• Two assessors were assigned per site. All the assessors were trained in a one day workshop. A total of fourteen expert clinicians were involved in the whole study (12 OT & 2 PT)

• In addition to repeated GRASSP testing, the International Standards of Neurological Classification for Spinal Cord Injury (ISCSCI) [2], Spinal Cord Independence Measure II (SCIM) [3], Capabilities of Upper Extremity Function Questionnaire (CUE) [4] were administered one time, Figure 1 defines the protocol and use of data for analysis.

• ANALYSIS: Guttman scaling to develop the scoring system; intraclass correlation coefficients (ICC) to establish reliability, Pearson correlation coefficients to establish validity with SCIM and CUE were conducted. Structural equation modeling was used to establish relationships between GRASSP subtests and function.

Figure 1. Reliability and Validity Protocol and Analysis



Results

• **Result 1:** Each subtest of the GRASSP (dorsal sensation, palmar sensation, strength, prehension ability and prehension performance) renders a total score. Therefore, five separate numeric values for each hand characterize the sensorimotor deficit of the upper limb. Table 1 and Figure 2 define how GRASSP scores are represented and interpreted.

• **Result 2:** Interrater and test retest reliability are above 0.8 for all subtests of the GRASSP. Reliability was calculated with ICC at a significance level of ($p<0.001$), see Table 2. Construct validity is demonstrated by greater sensitivity of the GRASSP sensation and strength tests when compared to the ISCSCI sensory and motor levels, see Figure 2. Concurrent validity is defined by the pearson correlation coefficients between GRASSP subtest total scores and the functional tests (SCIM, SCIM-SS, CUE), see Table 3.

• **Results 3:** Both palmar sensation and strength show significant relationships to prehension and upper limb function, see Figure 4.

Table 1. GRASSP Subtest Total Scores

ISCSCI	GRASSP Subtest Scores				
Sensory/ Motor/AIS Right Side	Dorsal Sensation 0 - 12	Palmar Sensation 0 - 12	Strength 0 - 50	Prehension Ability 0 - 12	Prehension Performance 0 - 30
C5/C4/A	4	3	5	0	0
C7/C6/A	6	9	23	10	21
C5/C6/D	10	10	26	5	16
C4/T1/D	12	12	45	12	27

Figure 2. GRASSP Subtest Total Scores

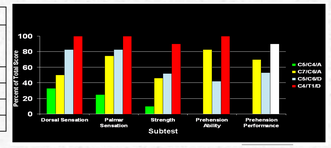


Table 2. Reliability of GRASSP Subtests

Subtest	Inter rater Reliability (n=72) ICC	Test Retest Reliability (n=45) ICC
Sensation	0.84* - 0.91*	0.86* - 0.95*
Strength	0.95* - 0.95*	0.98* - 0.98*
Prehension Ability	0.95* - 0.95*	0.98* - 0.98*
Prehension Performance	0.96* - 0.95*	0.93* - 0.96*

Table 3. Concurrent Validity

Subtest	SCIM	SCIM-SS	CUE
Sensation Total (R + L)	0.57*	0.74*	0.77*
Strength Total (R + L)	0.59*	0.74*	0.76*
Prehension Ability Total (R + L)	0.63*	0.77*	0.81*
Prehension Performance Total (R + L)	0.68*	0.79*	0.83*

Figure 3. Construct Validity – Agreement/Discordance of Sensory & Motor Items of GRASSP & ISCSCI

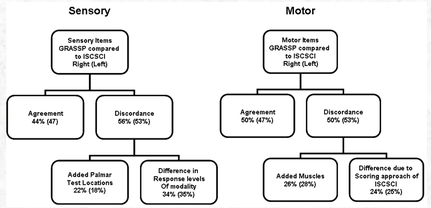
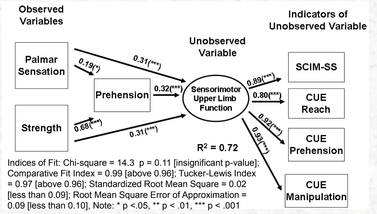


Figure 4. Relationships between GRASSP Impairment and Upper Limb Function



Indices of Fit: Chi-square = 14.3 p = 0.11 [insignificant p-value]; Comparative Fit Index = 0.99 [above 0.96]; Tucker-Lewis Index = 0.97 [above 0.96]; Standardized Root Mean Square = 0.02 [less than 0.09]; Root Mean Square Error of Approximation = 0.09 [less than 0.10], Note: * p <.05, ** p < .01, *** p < .001

Discussion

• This research has completed the development of the GRASSP, established the psychometric properties, and presented the relationships between impairment and function for individuals with tetraplegia.

• Some of the limitations of this work were that confounding impairments and their influence on the responses were not measured such as: neuropathic pain, peripheral neuropathy, effort and motivation.

• Responsiveness and response shift were not established for GRASSP.

• The GRASSP is a psychometric test and factors such as: response variation and human error can always be limitations of the measure.

• This work has however established some important factors in the field of measurement in SCI, such as:

- Confirming that sensation (palmar) plays a significant role in upper limb function for individuals with tetraplegia and should be measured, there is now evidence to support this
- Sensory and motor retraining can be targeted through interventions that represent sensorimotor function
- We have added a measure to the field which is novel reliable and valid.

• Future work planned for GRASSP development will be to establish responsiveness and minimally clinical important differences, define additional measures of sensation to be incorporated into GRASSP, establish GRASSP performance against other measures in the field, promote uptake for GRASSP in clinical practice for the assessment of SCI, and test for use in similar populations such as non traumatic SCI.

Conclusion

• In conclusion a new outcome measure has been added to the field of SCI which is specific to measuring impairment of the upper limb after tetraplegia and includes:

• A scoring approach established which defines impairment at the periphery with numeric values

• A method to summarize GRASSP scores

• Reliability is above 0.8 (ICC) for inter rater and test retest

• Construct validity shows the GRASSP has greater sensitivity than the ISCSCI

• Concurrent validity has been established with functional tests, and

• Sensory motor and sensorimotor function are important for the measurement of neurological status, particularly so that the connection between impairment and function can be made

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